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LIVER TRANSPLANTATION BEFORE ONE YEAR.
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Young infants are often refused for liver transplantation on the sole basis of young age. Over a total of 141 children who received an orthotopic liver transplantation in our centre (March 1984-July 1989), 17 patients (12%), were transplanted before they 1st birthday (15 biliary atresia, 1 Byler disease & 1 tyrosinemia). Mean age was 10,3 months (range 8-11) and mean weight 7,3 kg (range 5,2 - 13). A reduced liver was used 11 times over a total of 26 transplantations (42%). Immunosuppression included cyclosporine A, prednisone and azathioprine & OKT3 or ATG for steroid resistant rejection. Survivors were discharged after a mean hospital stay of 47 days (range 22-87) and the one year actuarial survival is 65% versus 77% in the whole serie. Reasons for retransplantation (29%) were primary non function (3), hepatic artery thrombosis (4) and rejection (2). 1 patient died perioperatively, 2 from primary non function, 1 from adenovirus infection, 2 from rejection & 1 from bone marrow aplasia. 18 rejection episodes, (11 corticoreistant) occurred in 11 patients. Liver transplantation can be proposed to young infants without age limits being imposed.

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MOSAIC PATTERN OF LACTASE EXPRESSION BY VILLUS ENTEROCYTES IN HUMAN ADULT-TYPE HYPOLACTASIA. L. Maiuri*, V. Raia*, J. Potter*, D. Swallow*, M. Wan Ho*, R. Fiocca*, G. Finzi*, M. Cornaggia*, C. Capella*, A. Quarani*, S. Auricchio*.

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Eight different monoclonal antibodies which recognize at least three distinct epitopes unique to the lactase protein of human enterocyte have been used to investigate lactase protein expression in hypolactasia of Neapolitan adults by light and electron microscopy with the immunogold technique. All the antibodies gave the same results, namely: strong brush border staining in all the 7 lactase persistent adults tested; no staining at all in 9 of the hypolactasia subjects and a mosaic pattern of staining of enterocytes in the other 12 adults with hypolactasia. The percentage of enterocytes showing intense staining for lactase protein varied between 4 and 20% of the total villus cells (mean value $50 \pm 9.86 \pm 5.84$). Intracellular staining was not apparent in any of the samples examined. Sucrase-isomaltase protein in contrast showed no mosaicism. The mosaic pattern of expression of lactase protein in some hypolactasic subjects suggests that columnar cells display a non homogeneous differentiation along the villus. If the two patterns, mosaicism or absence of detectable lactase protein, are present along the entire small intestine of the individuals tested, the results would suggest that two phenotypes of adult hypolactasia exist in the population we studied.

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MOSAICISM OF BLOOD GROUP SPECIFICITY OF BRUSH BORDER OF HUMAN ENTEROCYTES. S. Auricchio*, L. Maiuri*, V. Raia*, R. Fiocca*, E. Solcia*, G. Finzi*, M. Cornaggia*, Q. Norén*, H. Sjöström*, H. Skovbjerg*, D. Swallow*.

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The pattern of differentiation of the enterocyte along the villus is considered to be dependent on the age of the cell and uniform in cells occupying comparable location on the villus. Using a polyclonal antibody against A blood group specific components of enterocyte brush border and three monoclonal antibodies we have demonstrated the expression of the A and B antigens in the brush border and Golgi apparatus of enterocytes of individuals of the expected blood groups, by light and electron microscopy, with the immunogold technique. The polyclonal anti-A serum was specific for the brush border whereas the monoclonal reagents also bound the endothelial cells and erythrocytes in the sections. Four of 16 blood group A individuals showed a mosaic pattern of expression of the A antigen, both with the polyclonal serum and the monoclonal anti-A reagent. Some enterocytes showed a normal staining and some other enterocytes at similar position on the villus showed no staining at all. An anti-Le a antibody showed that all the 4 individuals with the mosaic pattern were non secretor. Abnormalities of blood group antigens may reflect abnormal expression of glucosyl-transferase genes or disturbances of their acceptors. Enterocytes may therefore display a non homogeneous pattern of differentiation in morphologically similar cells occupying comparable location on the villus.

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GENETIC STUDY OF HUMAN ADULT-TYPE HYPOLACTASIA BY ANALYSIS OF RESTRICTION FRAGMENT LENGTH POLYMORPHISMS (RFLP) OF THE LACTASE GENE. G. Sebastio, V. Guzzetta, B. De Vizia, A. Ballabio, W. Boll*, N. Mantei*, C. Somenza* and S. Auricchio.

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The adult-type hypolactasia is a condition due to homozygosity for a recessive allele of a hypothetical regulatory gene which controls the lactase activity in the adulthood. It is still unclear whether this regulatory gene is part of the lactase gene itself or maps to a different locus. To investigate this aspect, we have used a genetic approach: the analysis of family segregation of both the lactase gene and the lactase absorption capacity, which correlates with the lactase activity. 8 Italian families (20 meioses) entered this study. Families were chosen on the basis of the coexistence of subjects with either hypolactasia or persistence of high lactase activity. The lactase absorption capacity was assessed by breath-test after an oral load of 50 g of lactose. To follow the lactase gene segregation, we searched for RFLPs identified by the lactase cDNA. DNA of each subject was digested by the following restriction enzymes, which we found to detect RFLPs of the lactase gene: Pst I, Msp I, Rsa I, Bcl I. In all the informative pedigrees, cosegregation of the lactase gene and the hypolactasic condition has been observed. This result suggests that an event involving the lactase gene itself (e.g. regulation by sequences at the promoter; different RNA editing or splicing) can lead to either hypolactasia or, by mutation, to the persistence of high lactase activity. Indeed, we cannot exclude that hypolactasia might be a heterogeneous condition, in which also other mechanisms (e.g. post-translational modification) may control in trans the lactase gene expression.

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CHLORIDE SECRETION PATTERNS IN RECTAL EPITHELIUM OF CYSTIC FIBROSIS (CF) PATIENTS AND ITS RELATION TO THE $\Delta F508$ MUTATION. H. J. Veeze*, M. Sinaasappel*, D. J. J. Halley*, J. Bouquet*, J. Bijman*, H. R. de Jonge*.

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In CF the calcium (Ca) and cAMP mediated regulation of the chloride channel are defective in small intestine and proximal colon. In order to demonstrate the defect in rectal epithelium we studied suction biopsies in a modified Ussing chamber (exposed area 1.13 mm²) in 20 CF patients and 17 controls. Ca-mediated change in short circuit current (SCC) by Carbachol, positively associated with chloride secretion, was measured. The change in SCC $\pm$ S.E.M. (n) was $+24.5 \pm 3.1$ (17) in controls and -9.9 ± 2.6 (12) in CF (category I). Eight CF patients (including 3 sibs) showed a positive peak change in SCC of $+4.8 \pm 2.3$ indicating residual chloride secretion after the initial response of -5.2 ± 1.8 (category II). Homozygosity for the most common CF mutation ($\Delta F508$) was found exclusively in category I. Compound heterozygosity ($\Delta F508$ and another unknown mutation) was seen in 2 patients of category I and in all patients tested (n=6) for category II. CF patients with residual chloride secretion tend to have better lung functions and less disturbed sweat tests, not to be explained by differences in age. Reverse response in CF is a consistent and intriguing finding and represents a possible potassium secretion.

CONCLUSIONS: CF rectal epithelium of homozygotes for $\Delta F508$ showed an absent Ca-mediated chloride secretion corresponding with earlier studies of small intestine and proximal colon. One third of the CF patients however showed residual chloride secretion; these patients were heterozygotes for $\Delta F508$. In 2 compound heterozygotes without residual chloride secretion we suggest that the other mutation is of the same severity as $\Delta F508$.

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THE GENERATION OF A TOLERAGEN AFTER THE INGESTION OF OVALBUMIN IS TIME DEPENDENT AND UNRELATED TO SERUM LEVELS OF IMMUNOREACTIVE ANTIGEN

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During our research into the regulation of the gastrointestinal immune system (GALT) we have investigated the molecular and biological features of ovalbumin (OVA) which has been subjected to intestinal processing. Immunoreactive OVA absorbed by the gut was measured by a sandwich ELISA at different times after feeding 25mg OVA to adult mice. OVA was detected as early as 5 minutes after the feed ($36.7 \pm 16\text{ng/ml}$, mean $\pm$ 1SD) and reached maximal levels at 1 hour ($73.3 \pm 20\text{ng/ml}$). Pooled mouse serum, collected 5 minutes or 1 hour after the feed of OVA, was transferred i.p. into naive recipients. Suppression of systemic delayed hypersensitivity (DTH) was found in mice receiving 0.8ml of serum obtained 1 hour after OVA feeding but not when using serum obtained 5 minutes after feeding. In order to transfer serum samples containing similar levels of OVA, an increased amount (1.3ml) of serum collected 5 minutes post OVA feed was used in further experiments but again failed to induce DTH tolerance. Serum samples obtained 5 and 60 minutes after OVA feeding were analysed by fast protein liquid chromatography (FPLC) fractionation followed by ELISA. Both the charge characteristics and molecular weight of intestinally absorbed OVA were indistinguishable from native OVA. The results suggest that, although intact native OVA is the only molecular species detected by ELISA, it has no role in the suppression of DTH responses, and that the gut-processed tolerogen may be modified so that less than two antibody-binding epitopes remain.