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ARE SOLUBLE AND/OR MEMBRANE-BOUND TRANSGLUTAMINASE ACTIVITIES INVOLVED IN INTESTINAL METABOLISM OF GLIADINS?

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Transglutaminase (TGases) are Ca^{2+}-dependent enzymes, which link peptide-bound glutamine to primary amines. We found that wheat gliadins, purified A-gliadin and their derived peptic-tryptic peptides are effective acyl donor substrate for: 1) TGase purified from guinea pig liver 2) TGase activity of rat small intestine, measured both in jejunum slices and in influx chambers, in which only the jejunal mucosa was exposed to the substrates. The enzyme activity in jejunal mucosa homogenate, expressed as pmol of spermidine incorporated into N,N-dimethylated casein /mg/hour, is low at birth (0.10±0.02,mean±SD), increases up to 0.31±0.065 at 7-10 days and then decreases to the adult level (0.09±0.03) at 15 days of age. On the contrary, the enzyme activity of submucosa-serosa is from the 7th day at values similar to that of the adult intestine (0.5 and 0.8 respectively). 50% of TGase activity was detected in the particulate fraction (not containing brush border membranes) of both young and adult rat mucosa. These results are consistent with the hypothesis that TGase activities may be involved in the metabolism of gliadins, or of their peptides, in the lumen or in the mucosa. The specific cellular localization of the enzyme should be clarified.

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Intestinal permeability to macromolecules during viral enteritis in conventional and germ-free suckling mice. M. Heyman, G. Corthier, A. Petit, J.C. Meslin, C. Moreau, J.F. Desjeux. INSERM U.290 - Paris, INRA - Jouy-en-Josas, France.

Epithelial transport and degradation of horseradish peroxidase (HRP) was studied in conventional and germ-free suckling mice following an experimental infection with rotavirus. Conventional and germ-free mice developed diarrhea from days 2 to 8 post-infection (pi), with growth failure. Infectious virus detected by immunofluorescence was present in mucosal homogenates of germ-free mice up to day 8 pi, but persisted longer (day 13 pi) in conventional mice. Only mild histological lesions were associated with diarrhea but macrovacuolation and increased cellular density occured during the convalescence period (days 9 to 13 pi). Intact and degraded (HRP) fluxes from mucosa to serosa were measured in vitro, on pieces of jejunum mounted in Ussing chambers. A large increase in HRP permeability was noted during the initial diarrheal period (days 2 and 3 pi) in conventional mice and during the convalescence period (days 9 to 13 pi) in germ-free mice. In both cases, only intact-HRP transport rose -probably via a transcellular route- without alteration in degraded-HRP transport. These results indicate that rotavirus infection in mice causes an increase in intact HRP intestinal permeability which is dependent on intestinal microflora.

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EPIDEMIOLOGY AND CLINICAL FEATURES OF HEAT-STABLE ENTEROTOXIN PRODUCING E.COLI (STEC) IN ITALIAN CHILDREN WITH DIARRHEA SCREENED BY AN ELISA TEST

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Scanty information is available on ST-induced diarrhea in industrialized countries. We have screened for STEC 541 children (mean age 43 months, range 0-180 months) from 5 different Italian towns, with acute diarrhea using an Elisa method (1). This is the largest series of children screened for STEC in an industrialized country. All STEC strains were also testes by the suckling mouse assay (SMA) together with 90 strains negative in the Elisa. A total of 31 STEC (5.7%) were detected: 26 were positive in both tests, 4 only in the SMA, 1 only in the Elisa. The concordance was 96%, the sensitivity of Elisa was 87%. The highest rate of STEC isolation was found in spring and autumn. Mean age of children with STEC was 22 months, range 0-63 months. 48% of children with STEC had previously ingested potentially contaminated food, 16% had the previously contact with diarrheal subjects. Clinical features of children were dehydration (25%), fever (28%),vomiting (12%), abdominal pain (36%). In 36% of children other enteric pathogens were found associated with STEC. Values of fecal osmolarity and anion gap were consistent with a secretory type of diarrhea in 79% of children. Mean duration of diarrhea was 5 days. All but one patient did eventually well with oral rehydration therapy; one required parenteral rehydration. In conclusion: 1) the incidence of STEC induced diarrhea is higher in Italy than in other industrialized countries. 2) The Elisa test is a rapid and convenient tool to screen large numbers of strains. 3) Younger children are more susceptible to STEC-diarrhea then older children. 4)Clinical features are less severe in industrialized then in developing countries.

1. Thompson M. et al. J. Clin. Microbiol. 20, 59, 1984

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MICROFLORA AND BILE ACIDS IN PROTRACTED DIARRHEA AND COELIAC DISEASE Child Health Center, Warsaw,Poland

The aim of the study was to exam bacterial overgrowth/b.o./and bile acids/b.a./metabolism in children suffering from coeliac disease/CD/and protracted diarrhea/DP/. Material and methods :115 children suffering from CD and DP, of 1-36 month of age, have been examined.The children were divided into groups according to the b.o. or normal gut flora/n/:CDbo=27,CDn=19, DPbo=33, DPn=36.In duodenal fluid aerobes,anaerobes and yeasts were estimated.Serum b.a. were measured by RIA, duodenal b.a.-TLC,faecal b.a.-Sterognost 3-Pho. Results :Bacterial overgrowth was found in 47.8% in DP and 58.7% in CD, mainly caused by Enterobacteriaceae and among them E.Coli.E.Coli was isolated in 66.7% in DPbo and 60.7% in CDbo.In duodenal fluid unconjugated and secondary b.a. were found in 5% of children.In all the investigated groups malabsorption of b.a. was found-faecal b.a. over 270 umol/100g.In 34% fasting duodenal b.a. were below the critical micellar concentration/2mmol/l/.Fasting/0'/serum b.a. were elevated in 56% in DP and CD considered together and postprandialy/120'/ in 46%. Conclusions : 1. Bacterial overgrowth is a patogenic factor in DP and CD. 2. E.Coli in duodenal fluid cause prolongation and aggraviation of infant's diarrhea. 3. In both DP and CD bile acids malabsorption and liver failure are observed. 4. The estimation of microflora and b.a. give informations concerning prognosis and treatment among other MCT,but do not differentiate chronic diarrhea.

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EVALUATION OF ANTIGLIADIN IgA AS A SCREENING METHOD FOR COELIAC DISEASE IN CHILDREN.

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Serum IgA against gluten is a good marker of untreated coeliac disease, especially in children. In this study a new test (Pharmacia Gluten IgA EIA) was evaluated.
Assay was performed in microstrips which were gliadincoated.
Total test time: <3 h. The cut-off was established from 84 healthy children. It was determined to 25 arbitrary units (AU).
Study population: In total 91 children who had undergone a jejunal biopsy were studied. Of them 47 had villous atrophy and 44 had normal mucosa. 10 children with villous atrophy, from the previous group, treated with gluten-free diet. 19 children undergoing gluten challenge and biopsed the third time.
Results: Untreated children: All 47 children with villous atrophy had significant amounts of anti-gliadin IgA giving a sensitivity of 100%. 37 children out of 44 with normal mucosa had antibody levels <25 AU giving a specificity of 84%. The test gave a correct diagnosis in 92% of the cases.
After gluten-free diet: All treated children reacted with disappearance of anti-gliadin IgA (<25 AU).
After gluten-challenge: All children had anti-gliadin IgA above 25 AU.
Conclusion: All children with villous atrophy had significant antibody levels, which means that the test is a good screening method for candidates for jejunal biopgy. It is also a useful test for patient adherence to gluten-free diet.

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Anticholeraic effect and intestinal transepithelial passage of native and soluble formaldehyde-modified caseins. O. Kheroua, D. Tomé, D. Marcon-Genty, A. Ben Mansour, J.F. Desjeux. INSERM U.290, Hôpital Saint-Lazare, 75010 Paris, France.

An anticholeraic effect has been demonstrated for insoluble methylated total caseins in rat jejunum (Ped. Res. 18 : 1075- 1079, 1984). We studied the anticholeraic effect of native and soluble formaldehyde-modified β and κ purified caseins. Methods : Anticholeraic effect was determined in vivo in isolated rat jejunal loops. The transepithelial fluxes for intact proteins (ELISA) and degraded products (isotopic measurment) were performed in isolated stripped rabbit ileum in Ussing chamber in vitro. Results : Native β and κ caseins have no effect on cholera toxin induced secretion in vivo whereas the soluble formaldehyde-modified proteins significantly reduced this secretion. No intact transepithelial passage was detected for both native and modified β and κ caseins. The transepithelial fluxes of degraded products were for native and modified β-casein 1892 and 240 ng/h.cm^2 respectively, and for native and modified κ-casein 351 and 221 respectively. Conclusion : Formaldehyde-modified caseins have anticholeraic properties. This effect is probably due to an interaction of the modified proteins with the luminal side of the intestinal mucosa.