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Involvement of L-arginine/nitric oxide pathway in the absorption of calcium in the rat small intestine. R Schleiffer, M Galluser, F Raul (INSERM U61, Biologie Cellulaire et Physiopathologie Digestives, 3, avenue Molière, 67200 Strasbourg, France)

Nitric oxide (NO) is produced enzymatically in vivo from L-arginine. It was originally identified as the main endothelium-derived relaxant factor and is now recognized as a second messenger mediating a variety of biological processes, including platelet aggregation, neurotransmission and immunological reactions [Moncada et al (1991) Pharmacol Rev 43, 109-142]. Recent evidence indicates that enterocytes also produce NO [Blachier et al (1991) Biochem Biophys Acta 1092, 304-310]. The aim of this study was to determine the involvement of the L-arginine/NO pathway in intestinal calcium absorption.

An inhibitor of NO production (N<sup>ω</sup>-nitro-L-arginine methyl ester, l-NAME) and sodium nitroprusside (SNP), a donor of NO, were perfused in adult Wistar rats anesthetized with ethyl carbamate. An intestinal segment (duodenum + proximal jejunum) was perfused by both intraluminal and vascular routes [Scheiffer et al (1993) Biomed Pharmacother 47, 19-23]. The intraluminal solution (NaCl 155 mM + CaCl<sub>2</sub> 1.25 mM) contained 45Ca and was perfused at a flow rate of 0.2 ml·min<sup>−1</sup>. Calcium absorption was quantitated by measuring 45Ca appearing in the superior mesenteric vein. The drugs were intravenously perfused at a flow rate of 0.1 ml·min<sup>−1</sup>. Mesenteric blood flow (MBF) and mean arterial pressure (MAP) were also measured throughout the experiment. Perfusion of l-NAME (0.123 μmol·min<sup>−1</sup>) resulted in an increase in MAP (+17 mmHg) and a decrease in MBF (30%). Calcium absorption was decreased by 0.21 ± 0.02 to 0.15 ± 0.01 μmol·min<sup>−1</sup>·kg<sup>−1</sup> body weight within 20 min of l-NAME perfusion. Inversely, SNP (0.032 μmol·min<sup>−1</sup>) decreased MAP and increased MBF. An increase (30%) in calcium absorption was observed within 20 min of SNP perfusion.

These experiments suggest that the level of a product of the L-arginine/NO pathway may be involved in the regulation of calcium absorption in the rat small intestine.

Is gut permeability affected by age and soyabean antigenicity in the preruminant calf? P Branco Pardal 1, JP Lalles 1, F André 2, E Delval 3, R Toulec 1 (1 INRA, Laboratoire du Jeune Ruminant, 65, rue de Saint-Brieuc, 35042 Rennes, Cedex; 2 INSERM, Centre Hospitalier Lyon-Sud, Pierre-Bénite; 3 INRA, Station de Recherche sur la Nutrition des Herbivores, 63122 Saint-Genès-Champanelle, France)

Various small marker probes such as xylose, Cr-EDTA and mixtures of oligosaccharides are used to assess the permeability of the gut in health and disease, but the actual mechanisms of their uptake are still unclear. Dual marker tests like lactulose/mannitol [André et al (1990) Hepatogastroenterology 37 (suppl II), 113-117] rather than single probes are to be preferred to overcome extraneous influences, including gastric emptying rate or intestinal transit on marker absorption. Here, we assessed intestinal permeability using different markers in veal calves, which were fed milk replacers differing in protein source and antigenicity.

Twenty-eight male Holstein calves were placed in cages and fed various diets from 6 weeks of age until slaughter. Protein milk replacers was provided by either skim-milk pow-
der (SMP) or a mixture (40:60, CP basis) of whey and soyabean products including a hydrolysed isolate (HSPI), a concentrate (SPC) and a heated flour (HSF). The HSF was highly antigenic in vitro, while the others were not. Gut permeability measurements were carried out 1 week before (Po) and 3 (Pi) and 11 (P2) weeks after starting dietary treatments. Xylose (0.5 g/kg BW), Cr-EDTA (4 mg Cr/kg BW), and a mixture of sucrose (0.2 g/kg BW) and mannitol (0.1 g/kg BW) were fed separately as pulse doses. Calves were fasted 16 h before testing. Urine was collected quantitatively between 0 and 6 h post-dosing, and also up to 24 h for Cr-EDTA. Xylose was assayed colorimetrically, Cr by atomic absorption spectrometry, and oligosaccharides using gas-liquid chromatography [André et al (1990), op cit]. Marker excretion data (% dose) were analysed by non-parametric tests.

As expected, N digestibility was the highest with SMP, the lowest with HSF and intermediate with HSPI and SPC. Conversely, antisoya antibodies were detected in high levels with HSF only, confirming its high antigenicity. The age effect on the urinary excretion of markers was usually not significant, except for xylose excretion, which decreased between Pi and P2 in controls (33.8 ± SEM 2.92 vs 18.8 ± 5.72, P < 0.05). In that group, the sucrose/mannitol ratio also decreased (P < 0.05) from 1.27 ± 0.21 in P0 to 0.65 ± 0.09 in P1. Lastly, sucrose excretion also decreased (P < 0.05) from 2.73 ± 0.60 to 0.92 ± 0.28 between P0 and P2 with HSF. A significant diet effect was observed only in P1 using xylose and Cr-DTA. Their excretions were higher (P < 0.05) with SMP than with SPC or HSF (33.8 ± 2.92 vs 18.7 ± 3.61 or 16.0 ± 2.78) for xylose, and with SMP than with HSF (6.0 ± 0.99 vs 2.9 ± 0.37) for Cr-EDTA collected over 24 h. In conclusion, calf intestinal permeability was hardly influenced by diets, despite the low digestibility and high immunogenicity of HSF. Only xylose and Cr-DTA absorption were transiently reduced with some of the soya diets. A similar trend reported for xylose in calves fed soya products of unknown antigenic activity was interpreted as being due to an altered gut structure [Seegraber and Morill (1979) J Dairy Sci 62, 972-977]. This explanation does not fit for Cr-EDTA whose excretion should have increased in that situation. Finally, urinary excretion of Cr-DTA and oligosaccharides by calves differ substantially from those observed in humans [André et al (1990), op cit].

Systemic humoral responses to soyabean meal in dairy cows, their calves throughout weaning, and ruminating lambs. JP Lallès 1, RG Guilhermet 1, JL Troccon 2 (1 INRA-ENSA, Laboratoire du Jeune Ruminant, 65, rue de Saint-Brieuc, 35042 Rennes Cedex; 2 INRA, Station de Recherche sur la Vache Laitière, 35590 Saint-Gilles, France)

Soyabean meal (SBM) introduced in milk replacers for preruminant calves causes the development of severe gut disturbances and a high production of systemic antibodies (Abs) against dietary protein [Sissons (1982) Proc Nutr Soc 41, 53-61]. In contrast, ruminant calves appear to tolerate high levels of SBM, and produce only moderate levels of specific Abs. The systemic immune status of adult cattle and other ruminant species to dietary antigens has not been documented so far. Thus, we determined antisoya Ab titres in dairy cows, in their calves throughout weaning, and in ruminant lambs, all consuming SBM.

Jugular blood was collected from 14 dairy cows 2 months after calving, from their 14 calves after 2, 4 and 7 weeks of life, and from 14 ruminant lambs aged 2.5 months. Besides diets based on maize silage, cows consumed SBM from 1 kg/d 1 wk before calving to 3 ± 1 kg after, depending on milk production. Calves and lambs were weaned onto mixtures (80:20) of concentrates containing 17 and 12% SBM, respectively, and hay. Antisoya Abs were assayed in plasma by passive haemagglutination [Sissons (1982), op cit]. Ab titres (log2 dilution from initial dilution 1:20) were compared by non-parametric tests.

SBM usually contains 4-6% protein as near-native glycinin and β-conglycinin, which are highly immunogenic (eg, Ab titres of 8-12) in the preruminant calf [Sissons (1982), op cit]. Here, antisoya Ab titres were 1.29 (SEM 0.101) in cows, 0.00 (0.000), 1.36 (0.284) and 2.34 (0.291) in calves at 2, 4 and 7 weeks of age, respectively, and 1.00 (0.296) in lambs. Thus, the concentration of specific Abs in weaned calves was twice as high as in cows and lambs, and 400 times less than in preruminant calves fed antigenic soya [Sissons (1982), op cit]. This was probably linked with intense ruminal fermentation soon after weaning [Tukur et al (1993) Can J Anim Sci 73, 891-905]. However, titres increased significantly (P < 0.05 and P < 0.01) throughout weaning of calves, indicating the lack of systemic humoral