

Butyrate and glutamine metabolism in colonocytes: role of the intestinal microflora

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Conclusions:

- Part of ingested Propionibacteria survive digestive transit in a large number of subjects. They are not able to colonize the intestine, but promote the growth of colonic Bifidobacteria.
- Digestive motility, assessed by colonic transit time, is directly related to these bacterial changes.

Role of a human digestive strain of *Bac*teroides thetaiotaomicron in the metabolism of food-borne glucosinolates. L. Elfoul^a, S. Rabot^a, A.J. Duncan^b, L. Goddyn^b, N. Khelifa^c, A. Rimbault^c (^aInra UEPSD/MBS 78352 Jouy-en-Josas cedex; ^bMLURI Aberdeen, Faculté des sciences pharmaceutiques et biologiques; ^cLaboratoire de biologie/UMA Paris, France).

Consumption of brassica vegetables (cabbage, broccoli, etc.), which contain thioglucosides named glucosinolates, is associated with a lower risk of cancer. This property has been attributed to the isothiocyanates released by hydrolysis of parent glucosinolates under the action of plant myrosinase (EC 3.2.3.1). When the plant enzyme is inactivated by heating, the digestive microflora is responsible for the glucosinolate degradation into still unknown physiologically active compounds. Our aim was to investigate the ability of a strain of Bacteroides thetaiotaomicron, isolated from a human faecal microflora, to convert glucosinolates into isothiocyanates in vivo. Sixteen F344 germ-free rats were inoculated with the B. thetaiotaomicron strain and offered a glucosinolate-containing diet to simulate a realistic nutritional situation. They were dosed by stomach tube with 50 µmol of sinigrin, a pure glucosinolate commonly found in many brassica vegetables. Total

urine and faeces were thereafter collected separately over 48 h. In the faeces, excretion of intact sinigrin was quantified by HPLC and the release of its specific derivative, allylisothiocyanate, was detected by GC. Conversion of sinigrin into allylisothiocyanate was estimated by quantifying its major final metabolite, a specific urinary mercapturic acid, by HPLC. The B. thetaiotaomicron strain was able to degrade sinigrin since only 8 % of the oral dose was excreted intact in the faeces. Trace amounts of allylisothiocyanate were detected in the faeces 18 h after the administration of sinigrin and the mercapturic acids of allylisothiocyanate appeared in the urine as early as the 6th hour. Most mercapturic acid excretion occurred within 30 h following sinigrin administration and the proportion of sinigrin converted into allylisothiocyanate was estimated to be 13 %. This study shows that a strain belonging to the dominant human colonic microflora is able to degrade, in vivo, a glucosinolate commonly found in brassica vegetables, and to convert it into allylisothiocyanate, a compound with potential health benefits.

Butyrate and glutamine metabolism in colonocytes: role of the intestinal microflora. C. Cherbuy^a, C. Andrieux^b, C. Ide^a, C. Tuleu^c, M. Watford^a, P.H. Duée^a, B. Darcy-Vrillon^a (^aLNSA, Inra, 78352 Jouy-en-Josas cedex; ^bUEPSD, Inra, 78352 Jouy-en-Josas cedex; ³Lab. de Pharmacotechnie, Fac. des Sci. Pharm. et Biol. de Paris V, Paris, France).

The principal oxidative substrates of the colonic epithelium are butyrate, produced by bacterial fermentation, and circulating glutamine. We have previously shown that the capacity for ketogenesis from butyrate is lower, while the capacity for glutamine utilization is higher in the germfree rat. The goal of this study was to determine the role of the intestinal flora on the expression of the key mitochondrial enzymes, HMGCoA synthase and glutaminase.

Materials and methods: The following groups of rats (aged 3 months) were used: germfree (GF), conventional (CV), 2, 14 or 30 days after an inoculation of a conventional flora (IN) or 30 days after an inoculation of a flora that produces (*Clostridium paraputrificum*, CP) or does not produce (*Bifidobacterium breve*, BB) butyrate. Glutaminase activity was measured in isolated colonocytes. Colonic mucosa HMGCoA synthase was analysed by western and northern blotting.

Results: Activity of glutaminase was significantly lower in the group IN (30 days) when compared to germfree group (-55 %, P < 0.05) but similar to that of the conventional group. Similar results were obtained 2 days after inoculation. The levels of HMGCoA synthase mRNA and immunoreactive protein were similar in inoculated and conventional rats. Glutaminase activity in the BB group was not different from germfree animals, whereas CP rats had lower glutaminase activity, similar to that of conventional rats.

Conclusions: These results confirm that colonocyte butyrate and glutamine metabolism is controlled by the flora and show that the enzymes studied were valid markers of colonocyte metabolism. The rapid effect after inoculation implies a role for the production of bacterial metabolites, and comparison of CP and BB groups suggest that butyrate is involved. **Dietary fibre and enzymatic activity of the rabbit caecal flora.** T. Gidenne, V. Pinheiro (Station de recherches cunicoles, Inra, BP 27, 31326 Castanet-Tolosan, France)

An overly large reduction of the dietary fibre intake could lead to serious digestive troubles for the rabbit, which could be associated with a dysfunction of the caecal flora activity. The present study aimed to estimate the bacterial fibrolytic activity (BFA) for adult rabbits, fed ad libitum a control and a fibre deficient diet (NDF = 43.5 versus 26.4 %), and without changes in the fibre nature. The intake and the faecal digestibility were measured for 4 days, after a 10-day adaptation period. BFA was then measured on soft faeces according to the method of Jehl et al. (Jehl N., Gidenne T., Le Roux J.F., Proc. 6th World Rabbit Congress, 9-12 July, ASFC, 1996, vol. 1, pp. 199-203). Data (means and residual standard error 'SEM') were subjected to a monofactorial analysis of variance (table below).

A half reduction in the fibre intake resulted in a similar decrease in the pectinasic and cellulasic BFA, whereas xylanasic BFA and fibre digestibility were not significantly affected. Only pectinasic BFA was correlated ($r^2 = 0.58$; n = 14) with the quantity of fibre digested. Although the BFA was affected by a high inter-individual variability, this criterion seems relevant to appreciate the caecal flora activity according to the dietary fibre supply.

Diets	Fibre intake (g NDF/d)	BFA (nmoles red. sug. lib./g DM/h)			NDF digestibility	Fibre digested
		Cellulase	Xylanase	Pectinase	(%)	(g NDF/j)
Control $(n = 8)$	64.2	22	121	497	42.4	26.0
Deficient $(n = 8)$	35.8	.12	89	223	44.0	15.3
SEM	2.7	3	15	52	1.0	0.9
Stat. signif.: P	< 0.001	0.019	0.16	0.002	0.31	< 0.001