Lactose malabsorption and colonic fermentations alter host metabolism in rats

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Abstract

Lactose malabsorption is associated with rapid production of high levels of osmotic compounds, such as organic acids and SCFA in the colon, suspected to contribute to the onset of lactose intolerance. Adult rats are lactase deficient and the present study was conducted to evaluate *in vivo* the metabolic consequences of acute lactose ingestion, including host–microbiota interactions. Rats received diets of 25 % sucrose (S25 control group) or 25 % lactose (L25 experimental group). SCFA and lactic acid were quantified in intestinal contents and portal blood. Expression of SCFA transporter genes was quantified in the colonic mucosa. Carbohydrate oxidation (Cox) and lipid oxidation (Lox) were computed by indirect calorimetry. Measurements were performed over a maximum of 13 h. Time, diet and time × diet variables had significant effects on SCFA concentration in the caecum ($P < 0.001$, $P = 0.004$ and $P = 0.007$, respectively) and the portal blood ($P < 0.001$, $P = 0.04$ and $P < 0.001$, respectively). Concomitantly, expression of sodium monocarboxylate significantly increased in the colonic mucosa of the L25 group ($P = 0.003$ at $t = 6$ h and $P < 0.05$ at $t = 8$ h). During 5 h after the meal, the L25 group's changes in metabolic parameters (Cox, Lox) were significantly lower than those of the S25 group ($P = 0.02$). However, after 5 h, L25 Cox became greater than S25 ($P = 0.004$). Thus, enhanced production and absorption of SCFA support the metabolic changes observed in calorimetry. These results underline the consequences of acute lactose malabsorption and measured compensations occurring in the host's metabolism, presumably through the microbiota fermentations and microbiota–host interactions.

Key words: Lactose malabsorption: Colonic fermentation: Host–microbiota interactions: Indirect calorimetry

Lactose malabsorption affects 20 % of the population worldwide. It results from a decline in lactase expression, naturally occurring in mammals after weaning. This decline is within a specific time period for each animal species: 15 d for rabbits, 1 month for rats and to or until decades for human subjects[1,2]. A low intestinal lactase activity leads to the presence of undigested lactose in the large intestine, where it is then fermented by the microbiota. Lactose intolerance is defined as the onset of non-specific gastrointestinal symptoms such as bloating, rumbling, abdominal pain, nausea and diarrhoea. Although the factors responsible for lactose intolerance are not clearly understood, the intervention of osmotically active compounds is suspected[3]. Few treatments are available and many lactose-intolerant people are left with no other choice than to practise an exclusion diet that could lead to mineral and vitamin deficiencies.

The colonic metabolism of lactose has been reported to be associated with an increased production of bacterial metabolites, including SCFA (acetate, propionate and butyrate) and gases (hydrogen, carbon dioxide and methane)[4–7] as well as, under specific circumstances, organic acids (such as lactate, succinate and formate) and ethanol[8,9]. In physiological conditions (luminal pH 4·5–7), SCFA are specifically transported by monocarboxylate (MCT1) and sodium MCT1 (sMCT1) transporters present in the colonocyte brush border membranes[10,11]. Once absorbed, SCFA are carried in the portal vein to the liver where they can be oxidised. Butyrate is the preferred energy substrate of the mucosal cells[12,13]. In standard conditions, SCFA are at the forefront of the mammals' colonic health, and represent approximately 65 % of the colonic anions (70–130 mmol/l)[10–12]. However, their concentrations are 3-fold higher as a consequence of lactose

malabsorption[9,14]. Such metabolic modifications may be expected in sugar malabsorption. However, most experiments on lactose malabsorption have been conducted *ex vivo* or *in vitro*. Studying the *in vivo* kinetics of bacterial fermentation, colonic absorption and subsequent host metabolism is necessary in order to be able to determine the onset of lactose intolerance. Such data could also pave the way to new strategies to improve the clinical management of lactose intolerance.

Accordingly, the aim of the present study was to investigate, *in vivo*, the metabolic consequences of malabsorption after an acute ingestion of lactose in rats, and the consequences of host–microbiota interactions. For this purpose, the present study was carried out using the rat model in which the incomplete absorption of lactose and the subsequent overproduction by the gut microbiota of lactose-derived SCFA could be detected by indirect calorimetry. Rats were given a single meal of either 25% lactose or 25% sucrose. SCFA concentrations were quantified in the large-intestinal contents and portal blood, expressions of MCT1 and sMCT1 measured in the colonic mucosa and oxidation of carbohydrate (Cox) and lipid (Lox) recorded by indirect calorimetry. These measurements were performed over a maximal 13 h period.

Experimental methods

Animals and diets

The experiments were carried out following the European directive 2010/63/UE relative to the protection of animals used for scientific purposes, and were approved by the ethical committee of the National Institute of Agronomic Research (INRA) (approval reference 11/012). A total of eighty-four male Wistar rats (350–400 g) were housed under an artificial 12 h light–12 dark cycle (lights on at 08.00 hours). Being more than 12 weeks old at the onset of the studies, they were naturally lactase deficient. Rats were adapted for 8 d to a standard diet of the American Institute of Nutrition (AIN) of 74% carbohydrate (63% starch, 11% sucrose), 14% protein and 12% fat as energy. This standard diet was composed of dairy proteins (140 g/kg), starch (622 g/kg), sucrose (100 g/kg), cellulose (50 g/kg), soyabean oil (40 g/kg), mineral mix AIN-93M (35 g/kg), vitamin mix AIN-93 V (10 g/kg) and choline (2·3 g/kg)[15]. The experimental calibrated test meals were 5 g (362 kJ) of the standard diet modified to contain 25% (w/w) of either sucrose (control group, termed S25) or lactose (experimental group, termed L25) added at the expense of starch. The rats were randomly assigned to one of the following groups: L25 or S25. The test meals were administered after an overnight fast. $n_{i,j,k}$ = seventy-two rats were serially killed for sample collection, where: i(diet) = 1,2 (1 = L25, 2 = S25), j(time point from diet ingestion) = 1,…,6 (1 = 0, 2 = 2 h, 3 = 4 h, 4 = 6 h, 5 = 8 h, 6 = 13 h) and k(rat) = 1,…,6. $n_{i,k}$ = twelve rats were assigned to the indirect calorimetry study.

Blood, intestinal contents and colonic mucosa sampling and SCFA extraction

Portal blood (2 ml) was sampled directly from the hepatic vein using a heparinised syringe. Blood samples were centrifuged for 4 min at 4°C and 600 g. A volume of 315 μl of the plasma fraction was kept in 35 μl of 5% H_3PO_4 for derivatisation and GC quantification. The rest of the plasma fraction was stored at −80°C until analysis. The colonic tissues were washed in ice-cold 0·9% (w/v) NaCl (pH 7·0) and the mucosa was scraped off using a glass slide before immediate freezing in liquid N_2 and storage at −80°C until quantitative real-time-PCR analyses. The caecal and proximal colonic contents were simply sampled in sterile tubes. SCFA were extracted by vigorous mixing with deionised water. The volume of deionised water added was 0·9-fold of the weight for caecal samples and 1·8-fold for colonic ones. As with the plasma fraction, 315 μl of the supernatant were mixed with 35 μl of 5% H_3PO_4 for subsequent derivatisation and GC quantification. The rest of the caecal and colonic fractions were kept for enzymatic reactions and spectrophotometric quantification. Samples were stored at −80°C until further analysis.

SCFA quantification

The SCFA were measured after derivatisation (esterification) with a modified Kristensen technique[16], followed by GC. This method is based on SCFA esterification by 2-chloroethyl-chloroformate in aqueous and acid media, using isocaproic acid as the internal standard. More precisely, 350 μl samples (315 μl sample + 35 μl 5% H_3PO_4), prepared as mentioned earlier, were mixed with 20 μl of 37% HCl and 100 μl of 1% isocaproate solution (w/v). Then, 750 μl of acetonitrile, 750 μl of 2-chloroethanol and 100 μl of pyridine were added. After careful homogenisation, the mixtures were centrifuged for 10 min at 3000 rpm and 4°C. The reaction was initiated by adding 50 μl of 2-chloroethyl chloroformate to 1·5 ml of the supernatant. Subsequently, 4 ml of distilled water were added and the esters were extracted with 500 μl of cyclohexane. They were then analysed with a GC (Varian CP3800; Varian Medical Systems) equipped with a capillary column (30 m, 0·32 mm inner diameter, Restek Rtx 502.2; Restek) and fitted with a flame ionisation detector and a split/splitless injector. Helium was used as the carrier gas (1 ml/min) and 6 μl of each sample were injected. The column was maintained at 75°C for 6 s, then the temperature was increased to 170°C at a rate of 7°C/min. This temperature was maintained for 10 min, before being increased to 220°C (50°C/min) and maintained for 3 min. The injector and the detector were utilised at 200 and 300°C, respectively. The amount of SCFA was determined with reference to an internal standard (isocaproate) and external standards (200 mM-acetate, 80 mM-propionate, 80 mM-butyrate, 10 mM-isobutyrate, 10 mM-isovalerate, 8 mM-valerate and 8 mM-caproate).

Lactate enzymatic assay

D/L-Lactate concentrations were determined with a commercial kit (Megazyme International Ireland Limited). Samples, prepared as mentioned earlier, were homogenised and centrifuged for 5 min at 4°C and 14 000 g. The supernatant was collected for a second round of centrifugation for 15 min at

4°C and 14 000 g. A volume of 500 µl of the supernatant was applied to a spin column of a centrifuge nanosep filter (30 kD; Pall Corporation) and centrifuged at 4°C and 14 000 g, until filtration of the whole volume. D/L-Lactate was then quantified in the final ultra-filtrates, according to the manufacturer's recommendations.

Gene expression of sodium monocarboxylate and monocarboxylate

The colonic expression of genes solute carrier family 16, member 1 (*SLC16A1*) and solute carrier family 5 (iodide transporter), member 8 (*SLC5A8*), which, respectively, encode for the MCT1 and sMCT1 transporters, was investigated using quantitative RT-PCR. The colonic mucosa was scraped and total RNA isolated using TRIzol reagent, according to the manufacturer's protocol (Ambion/Applied Biosystems, Life Technologies Corporation). Sample quality was assessed by 260/280 nm absorption ratio determination (between 1·8 and 2·0) and checked on a 1% agarose gel. For quantitative RT-PCR analyses, 1 µg total RNA was first reverse transcribed using the High Capacity cDNA Archive kit (Applied Biosystems). Reverse transcribed RNA was amplified on a thermal cycler (7300 Real-Time PCR System, Applied Biosystems) using the SYBR green (Power SYBR Green, Applied Biosystems) fluorescence method and specific oligonucleotides. The primers used were as follows: rat RPL13A – forward 5′-GGATCCCTCCACCCTATGACA-3′, reverse 5′-CTGGTACTTCCACCCGACCTC-3′; rat MCT1 – forward 5′-GCAACGACCAGTGAAGTGT-3′, reverse 5′-AAGCCCAAGACCTCCAAT-3′; and rat SMCT1 – forward 5′-CT GGGCTTGTTTTCTTTGG-3′, reverse 5′-CGTTGTGCGTGCTGTTAC-3′. Results were analysed with 7300 SDS system software (Applied Biosystems). Quantification of RNA was carried out by comparison of the number of cycles required to reach reference and target threshold values ($\delta - \delta C_t$ method).

Indirect calorimetry

Cox and Lox were computed using the Weir equation from continuous measurements of V_{O2} and carbon dioxide production (V_{CO2}) by indirect calorimetry[17]. The metabolic cage was equipped with force transducers placed below the metabolic chamber. This made possible the removal of the part of the respiratory exchange that was directly related to the energy expended on spontaneous activity. Thus, it was possible to compute meal-induced changes in Cox and Lox, free of short-term changes due to the cost of activity[18]. The rats were housed in the metabolic cage at 18.00 hours, with free access to water but no access to food. Temperature in the cage was regulated at 26°C to limit energy expenditure for thermoregulation. Basal Cox and Lox were measured between 06.00 and 10.00 hours the next day. The test meals were given at 10.00 hours and the changes in Cox and Lox *v.* baseline were measured during 8 and 13 h. At the end of the experiment, rats were euthanised by an intra-peritoneal injection of pentobarbital (0·1 ml/100 g body weight) and the intestinal contents, the intestinal mucosa and the portal blood samples were collected.

Statistical analyses

Results are presented as means with their standard errors. Analyses were performed with either an ANOVA using SAS software (version 9.1, SAS Institute, Inc.) or a Student's *t* test for the unpaired data.

Results

SCFA and lactic acid quantification in intestinal contents and portal blood

Lactic acid and SCFA were quantified in intestinal contents and portal blood in L25 (25% lactose diet) and S25 (25% sucrose diet) groups (Fig. 1 and Table 1). An increase of SCFA concentration was observed in intestinal contents
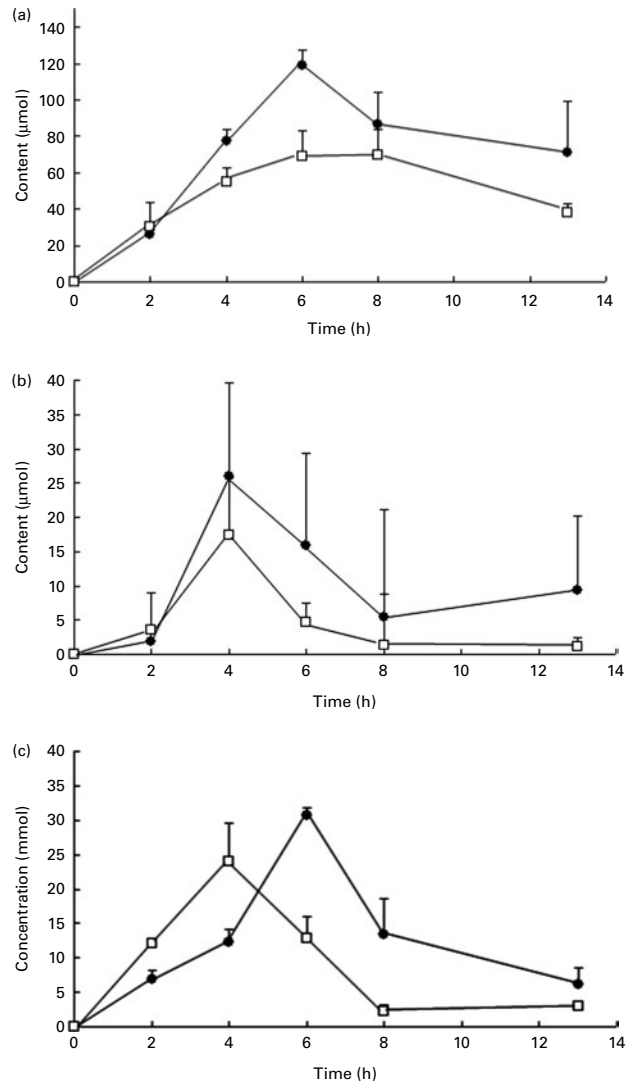

Fig. 1. Total SCFA variations after L25 (●) and S25 (□) diets in (a) caecum, (b) proximal colon, (c) portal blood. S25: diet containing 25% sucrose; L25: diet containing 25% lactose. Time ((a) $P<0.001$, (b) $P=0.24$, (c) $P<0.001$), diet ((a) $P=0.004$, (b) $P=0.06$, (c) $P=0.04$) and time × diet ((a) $P=0.007$; (b) $P=0.63$; (c) $P<0.001$) effects were evaluated by ANOVA. A *P* value over 0·05 is considered as not significant. Comparisons between S25 and L25 were performed at each time point by a Student's *t* test for unpaired data and are presented in Supplementary material (available online). $n_{i,j,k} = 72$, where i(diet) = 1,2, j(time point from diet ingestion) = 1,…,6 and k(rat) = 1,…,6.

Table 1. Acetate, propionate, butyrate and lactate in the caecum, proximal colon and portal blood after L25 and S25 diets*

(Mean values with their standard errors)

	S25	L25	SEM	P
Caecum				
Acetate	0·52	0·76	0·02	0·001
Propionate	0·11	0·14	0·01	0·17
Butyrate	0·04	0·05	0·003	0·54
Lactate	0·23	3·96	0·16	0·001
Proximal colon				
Acetate	0·12	0·18	0·01	0·21
Propionate	0·01	0·03	0·001	0·01
Butyrate	0·005	0·04	0·01	0·54
Lactate	ND	1·29	0·06	–
Portal blood				
Acetate	59·6	95·6	3·07	0·01
Propionate	47·7	56·8	3·35	0·47
Butyrate	16·8	22·6	1·67	0·29
Lactate	0·07	0·08	0·003	0·56

S25, containing 25 % sucrose; L25, containing 25 % lactose; ND, not detected.

* Data are expressed as means of AUC from the plot of contents (μmol) $v.$ time (h) ± pooled SEM for caecum and colon, and concentration (mmol/l) $v.$ time (h) ± pooled SEM for portal blood. Data are compared by a Student's t test for unpaired data. A P value over 0·05 is considered as not significant. $n_{i,j,k} = 72$, where i(diet) = 1,2, j(time point from diet ingestion) = 1,...,6 and k(rat) = 1,...,6.

(caecum and colon) for both groups, with a peak occurring between 4 and 8 h after the ingestion of the test meal. The higher amounts of SCFA and lactic acid were mainly encountered in the caecum. Total SCFA were 140 % higher in the caecum and 80 % higher in the colon in the L25 group compared with the S25 group. Time, diet and time × diet variables presented significant effects on caecal concentrations of SCFA ($P<0·001$, $P=0·004$ and $P=0·007$, respectively) and lactate ($P=0·004$, $P<0·001$ and $P=0·001$, respectively). Significant differences, presented in the Supplementary material (available online), were especially noted at 6 h ($P=0·004$) after the ingestion of the test meal. No significant difference was noticed in the proximal colon between the L25 and S25 groups, except for propionate concentrations ($P<0·05$). A moderate increase of SCFA was also noticed after the S25 meal. Finally, quantifications in the portal blood also revealed an increase of total SCFA over time in the S25 and L25 groups, with peaks occurring at 4 and 6 h after the diets, respectively. Time, diet and time × diet variables presented significant effects on SCFA concentrations in the portal blood ($P<0·001$, $P=0·04$ and $P<0·001$, respectively). Significant differences, presented in the Supplementary material (available online), were especially noted at 4 h ($P=0·009$) and 6 h ($P<0·001$) after the ingestion of the test meal. Total SCFA concentrations exhibited a 2-fold higher peak in the L25 group compared with the S25 group, probably driven by a significant increase of acetate concentrations ($P=0·01$) (Table 1).

Expression of sodium monocarboxylate and monocarboxylate genes in the colonic mucosa

The expression of SCFA transporters sMCT1 and MCT1 was analysed in the colonic mucosa in order to determine their absorption. Quantitative RT-PCR analyses revealed that diet presented a significant effect on the expression of sMCT1 ($P=0·02$) (Fig. 2). The differences, presented in the Supplementary material (available online), were significant for sMCT1 at 6 h ($P=0·03$) and 8 h ($P<0·05$).

Indirect calorimetry after a sucrose- or lactose-containing test meal

Calorimetric records following ingestion of the test meals revealed that Cox and Lox, respectively, presented a significantly lower increase and lower inhibition after ingestion of an L25 meal compared with an S25 meal (Fig. 3). Time, diet and time × diet variables presented significant effects on Lox ($P<0·001$, $P<0·001$, $P=0·004$, respectively) and Cox ($P<0·001$, $P<0·001$, $P<0·001$, respectively) kinetics over the 13 h post-ingestion. During the 8 h period after the test meals, the difference in Cox between the L25 and S25 groups was 6·6 kJ (Table 2). Interestingly, the differential amount (L25−S25) of total SCFA accumulated in the caecum resulted in a differential energy of 7 kJ (difference of 2443 μmol, with an average of $M = 74$ g/mol). After intersection of the curves, a recovery of energy occurred for the L25 group, but remained partial
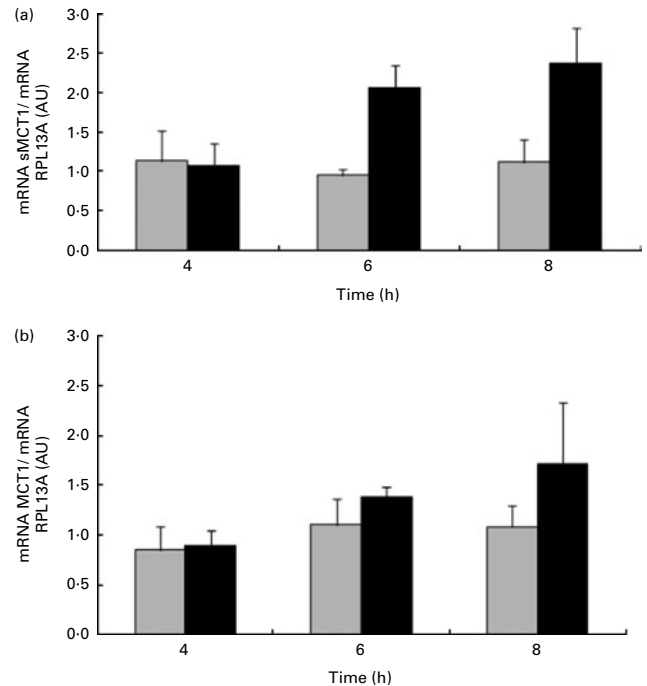

Fig. 2. (a) Sodium monocarboxylate (sMCT1) expression after L25 (▬) and S25 (▬) diets; (b) monocarboxylate (MCT1) expression after L25 and S25 diets. S25, diets containing 25 % sucrose; L25, diets containing 25 % lactose; Time ((a) $P = 0·24$, (b) $P=0·34$), diet ((a) $P=0·02$, (b) $P=0·29$) and time × diet ((a) $P=0·19$, (b) $P=0·72$) effects were evaluated by ANOVA. A P value over 0·05 is considered as not significant. Comparisons between S25 and L25 were performed at each time point by a Student's t test for unpaired data and are presented in Supplementary material (available online). $n_{i,j,k} = 36$, where i(diet) = 1,2, j(time point from diet ingestion) = 3,4,5 and k(rat) = 1,...,6. RPL13a, ribosomal protein L13a; AU, arbitrary units.
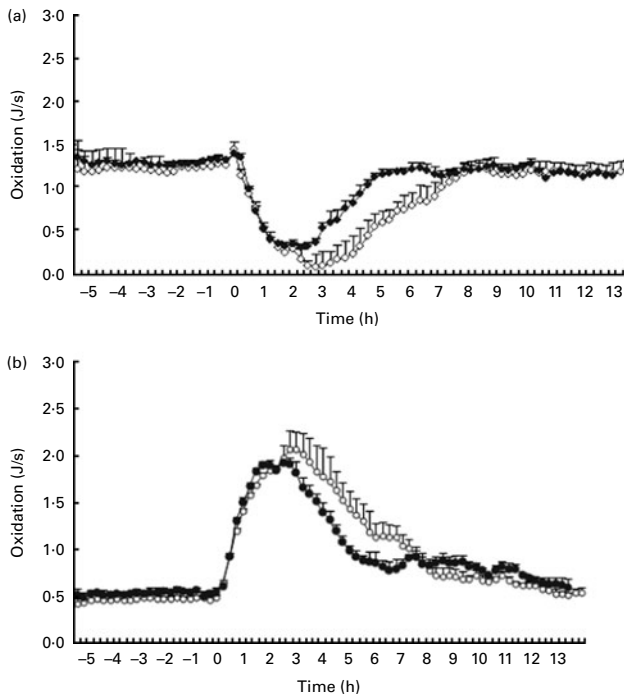
Fig. 3. (a) Lipid oxidation and (b) carbohydrate oxidation kinetics over 13 h after L25 (●) and S25 (○) diet. S25: diet containing 25 % sucrose; L25: diet containing 25 % lactose. Time ((a) $P < 0.001$, (b) $P < 0.001$), diet ((a) $P < 0.001$, (b) $P < 0.001$) and time × diet effects ((a) $P = 0.004$; (b) $P < 0.001$) are evaluated by ANOVA. A P value over 0.05 is considered as not significant. $n_{i,k} = 12$, where i(diet) = 1,2 and k(rat) = 1,...,6.

with a difference of less than 2·5 kJ between the 8 and 13 h. As the decrease in Cox was compensated by a lower inhibition of Lox, overall postprandial thermogenesis (Cox + Lox) was not significantly different between the L25 and S25 groups.

Discussion

The present study contributes to a better knowledge of the *in vivo* kinetics of the metabolic consequences of acute lactose

Table 2. Meal-induced changes (kJ) in carbohydrate oxidation (Cox) and lipid oxidation (Lox) during the periods 0–8 h and 8–13 h, after single L25 and S25 meals*

(Mean values with their standard errors)

	S25		L25		
	Mean	SEM	Mean	SEM	P
0–8 h					
Cox	40·6	2·5	34·0	1·8	0·049
Lox	16·7	1·8	25·1	1·0	0·009
8–13 h					
Cox	11·5	0·4	13·9	0·8	0·004
Lox	21·0	2·0	21·3	0·3	0·88

S25, containing 25 % sucrose; L25, containing 25 % lactose.
* Data are expressed as means of Cox or Lox plots (kJ) ± SEM and are compared by a Student's t test for unpaired data. A P value over 0·05 is considered as not significant. $n_{i,k} = 12$, where i(diet) = 1,2 and k(rat) = 1,...,6.

malabsorption in rats. Lactose malabsorption appears to be mainly associated with modifications in the production of SCFA by the gut microbiota and their subsequent absorption and metabolic utilisation by the host. The rat model provides control of both internal parameters, such as genetic background (allowing similar physiological responses), and external parameters, such as the 13 h kinetics in the calorimetric chamber plus samplings of the portal vein, the intestinal contents and the colonic mucosa.

Lactose malabsorption led to microbial production (quantified in intestinal contents) and colonic absorption (indicated by quantification in portal blood and sMCT1 expression in the colonic mucosa) of SCFA. Finally, the consequent metabolic changes occurring in the lactose malabsorber host were supported by the calorimetric parameters.

In the mammalian colon, a close similarity in the molar ratio of acetate, propionate and butyrate is noted among species. The SCFA profiles are not fixed in human subjects, but, in standard conditions, the molar ratio has been reported to be about 60:20:20 (acetate:propionate:butyrate) in colonic contents and about 85:10:5 in blood, with some variation depending upon intestinal segment (left or right) and blood sample origin (portal, hepatic or peripheral)[19]. Indeed, these molar ratios may be altered by consumption of fermentable fibres or modified foods. Recent studies also underline the role of dietary proteins in the SCFA production, either in quantity or quality[20,21]. Such questions are of interest for the medical community, as SCFA, and especially butyrate, have demonstrated to have a beneficial effect on the host. The main health benefits attributed to butyrate are its role in the reduction of colonic inflammation and permeability[22] and in the protection against colorectal cancer[23]. SCFA concentration in the large bowel is the result of SCFA production by the intestinal microbiota, of *in situ* metabolisation by colonocytes and of colonic absorption. Therefore, high SCFA concentrations in the caecum may be explained either by an active production and/or by a low uptake. Furthermore, once absorbed, SCFA are carried through portal blood to the liver, where most of them are metabolised, accounting for the consecutive lower concentrations in the peripheral blood[19]. In the present study, after ingestion of lactose, lactic acid and total SCFA were significantly higher in the caecum and the portal blood. One may assume that these differences account for lactose ingestion specifically, as L25 and S25 groups had both the same protein intake. Interestingly, major differences were noticed 6 h after lactose ingestion. The bioavailability of substrate and fermentation kinetics of the microbiota may explain the delayed caecal production of intermediary and end metabolites after the ingestion of lactose[24]. The substantial absorption of colonic SCFA, usually between 6·1 and 12·6 μmol/cm² per h[25], explains the gradual metabolite concentration decrease and pH increase (from 5·6 to 6·6) from the caecum to the distal colon[19]. These findings could account for the absence of significant differences in the proximal colon after S25 or L25 diets. Similar kinetics in bell curves were observed for SCFA variations in the caecum, the proximal colon and the portal blood. However, the peaks were observed at 6 h after

the L25 meal in the caecum and the portal blood, and at 4 h after the meal in the proximal colon.

The decrease after 4 h of SCFA colonic concentrations is concomitant to the increase of mRNA for sMCT1 and MCT1 transporters in the colonic mucosa. This observation could suggest that, before 6 h, the activity of these transporters was disrupted (pH drop) or exceeded (unusually high concentrations of SCFA), leading to SCFA accumulation until the biosynthesis of a new pool of transporters. The pattern of the SCFA concentrations in portal blood is consistent with this hypothesis. We did not follow-up the transcription of sMCT1 and MCT1 in the caecal mucosa, but *in vitro* experiments proved that SCFA abundance resulted in MCT1 and sMCT1 mRNA rises, probably in order to maximise intracellular availability of SCFA[26–29]. An increase in passive uptake through the paracellular route may be also implied in the increased SCFA concentrations in the portal blood, as a fructo-oligosaccharide-induced increase in intestinal permeability was demonstrated in rats[30].

At last, indirect calorimetry underlines the smaller increase of Cox, the smaller inhibition of Lox and the fairly stable overall thermogenic response after ingestion of an L25 meal compared with an S25 meal. Indirect calorimetry confirms the lower absorption and subsequent metabolic utilisation of lactose in rats, with a reversed trend between the L25 and S25 groups, beyond 6 h after the test meals. Finally, with the L25 diet, the excess energy available in caecal contents (as SCFA) is in the same range (6–7 kJ) as the energy underexpended for Cox during 8 h after the ingestion of the diet. Furthermore, the recovery of this amount of energy may also explain the absence of differences when comparing the thermogenesis after lactose and sucrose diets. These results can be interpreted as the oxidation of the SCFA produced by the microbial fermentation of lactose, whose metabolites are then absorbed and oxidised by the host. Thus, the use of new metabolites, largely produced and accumulated under lactose ingestion, may be considered as capable of providing this delayed energy. Such an idea has already been suggested in a calorimetric study in rats fed with various dietary fibres, in which Cox remained higher for a longer period after the ingestion of the fibres than after the ingestion of the control diet[31]. Among the accumulated metabolites, caecal and colonic SCFA are presumably playing a major metabolic role. Indeed, several studies confirm their capacity to be easily absorbed across the colonic mucosa, resulting in a salvage of carbohydrate energy that otherwise would have been lost in faeces[32–34].

Overall, the results provided by the measurements of the luminal and the blood SCFA, transporter gene expression and by indirect calorimetry method showed that up to a given load of undigested lactose, metabolic changes occur from 6 h after lactose ingestion. They involve caecal and colonic fermentation of lactose, leading to the caecal production of SCFA, subsequent activation of SCFA-specific transporters and colonic absorption of the SCFA that are metabolised in the host's tissues, accounting for the metabolic compensations from 6 h. The present study emphasises the adaptation of both the colonic microbiota and the host metabolisms, but also supports the suggested nutritional strategies to optimise the microflora and host adaptation in order to alleviate the symptoms of lactose intolerance. These results raise the question of the metabolic reorientation of the colonic microbiota and the host over chronic lactose ingestion by lactose malabsorbers. They also pave the way to the use of indirect calorimetry as a non-invasive tool to detect intestinal lactose malabsorption and to explain, at least partly, the subsequent metabolic adaptation through microbial fermentations. Clinical trials are needed to confirm the validity of this methodological approach in human subjects.

Supplementary material

To view supplementary material for this article, please visit http://dx.doi.org/10.1017/S0007114512005557

Acknowledgements

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