Dental and metabolic effects of lactitol in the diet of laboratory rats

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1. Because so little is known about the properties of lactitol as a possible alternative bulk sweetener to sucrose, it was tested in two large-scale experiments in laboratory rats. Matched groups of caries-active Osborne–Mendel rats were fed on uniform diets containing lactitol and compared with a sucrose control in both experiments, plus a xylitol control in the first experiment.

2. In the early stages of the experiments weight gains and food utilization were better on the sucrose than on the lactitol regimens. Body-fat storage was higher on the sucrose than on the polyol regimens.

3. At the end of 8 weeks the mandibular molars were examined for dental plaque accumulation and dental caries. The dental caries scores when 160 g sucrose/kg in the diet was replaced by lactitol were lower by a highly significant margin, bringing them down to the same low level as those on a 160 g xylitol/kg regimen.

4. Testing lactitol in a manufactured food product, shortbread biscuits, in comparison with ordinary sucrose biscuits, showed differences in plaque scores (significant) and caries levels (highly significant), with 60% fewer lesions on the lactitol regimen.

5. The results confirm the low cariogenic potential of lactitol, but show metabolic differences compared with sucrose.

The polyols put forward as bulk sweeteners to replace sucrose in foods and drinks are mostly monosaccharide derivatives, e.g. sorbitol, mannitol and xylitol, and very little attention has been given to the use of disaccharide polyols. One of these is lactitol, made by the hydrogenation of lactose. Lactitol is said to have good properties for food manufacture (den Uyl, 1987), but its dental and metabolic properties have not been investigated in any detail. In microbiological fermentation studies in vitro, acid production from it was slow but increased on adaptation of *Streptococcus mutans* (Havenaar *et al.* 1978), and acid development, enamel demineralization and microbial polysaccharide formation were all low from lactitol in comparison with five other bulk sweeteners (Grenby *et al.* 1989). In a single trial in man, sweets made with lactitol instead of sucrose reduced the amount of plaque on the teeth and altered its composition (Grenby & Desai, 1988).

Experimental work in animals is also lacking. In just one short report van der Hoeven (1986) observed that substituting lactitol for sucrose at 250 g/kg diet for 3 weeks significantly diminished the caries increment in programme-fed rats. The object of the experiments reported here was to take this a stage further, using a longer period of feeding, initially comparing lactitol with a positive control (sucrose) and a negative control (xylitol) at a level of 160 g/kg diet, which approximates to the average dietary sucrose content in developed countries, and then progressing to test lactitol as a replacement for sucrose in a manufactured food known to be highly cariogenic in this experimental system, sweet biscuits (Grenby & Paterson, 1972; Grenby & Bull, 1977; Grenby, 1987). Not only the dental status of the animals, but also their food and water intakes, weight gains, body-fat storage and energy retention were monitored in the first of the two experiments, in order to provide information on toleration of lactitol and xylitol, and their metabolism compared with sucrose.

Table 1. *Expt 1. Composition of diets (g/kg)*

Dietary regimen ...	Lactitol	Xylitol	Sucrose
Lactitol monohydrate	160	—	—
Xylitol	—	160	—
Sucrose (caster sugar)	—	—	160
White flour	497·5	497·5	497·5
Skim-milk powder	320	320	320
Desiccated liver powder	20	20	20
Vitamin–mineral–EFA supplement*	2·5	2·5	2·5

EFA, essential fatty acids.
* SA-137; Intervet Laboratories Ltd, Cambridge.

Table 2. *Expt 2. Composition of powdered diets (g/kg)*

Shortbread biscuits containing 166 g/kg of: Lactitol	660	—
Sucrose	—	660
Skim-milk powder	320	320
Desiccated liver powder	20	20
Vitamin–mineral–EFA supplement*	2·5	2·5

EFA, essential fatty acids.
* SA-37; Intervet Laboratories Ltd, Cambridge.

MATERIALS AND METHODS

Animals

Litters of a caries-active strain of Osborne–Mendel rats were divided at weaning at 21–24 d of age into matched groups and fed on the test diets for the next 8 weeks. They were kept on screen-floor cages to prevent access to bedding, etc. and no attempt was made to alter their natural oral microflora. They were examined regularly (daily at first) for any signs of debility, loss of condition, diarrhoea and behavioural changes. They were weighed every 2 weeks, and a complete record was kept of food and water consumption.

Dental examination

The methods are given in full by Grenby (1988). Plaque was measured on the mandibular molars on a 0–3 scale, and the values were added to give a plaque score for each animal. Caries scoring was based on the method of Shaw *et al.* (1944), grinding down the teeth from the occlusal surface to evaluate fissure, interproximal and smooth-surface lesions under twenty-five times magnification.

Carcass analysis

Samples of homogenized body tissues were dried to constant weight. Total lipids were measured by the method of Folch *et al.* (1957), protein by the small-scale Kjeldahl method, and energy value by ballistic bomb calorimetry.

Statistics

The main findings are given as means with their standard errors. Inter-group comparisons were made by Student's t or the F test, as appropriate.

Table 3. *Summary of water and food intake values and weight gains of rats receiving diets containing lactitol (L), xylitol (X) or sucrose (S)**

	Males			Females		
Dietary regimen...	L	X	S	L	X	S
Mean water intake (ml/rat per d)						
0–14 d	16·2	15·9	11·0	13·2	13·3	9·7
15–28 d	17·7	26·0	17·6	15·9	14·3	17·3
29–42 d	24·4	29·9	23·0	22·0	28·1	20·6
43–56 d	14·9	18·7	17·4	16·4	14·5	16·3
Mean food intake (g/rat per d)						
0–14 d	6·8	6·4	10·4	6·4	4·7	10·1
15–28 d	8·7	—	10·9	7·9	—	9·8
29–42 d	14·2	16·9	17·6	11·5	17·9	14·0
43–56 d	20·6	21·4	20·7	18·3	17·7	17·6
Wt gain (g/rat)						
0–14 d						
Mean	43	29	66	36	23	55
SE	1	4	1	1	2	2
15–38 d						
Mean	65	67	68	46	45	38
SE	2	3	1	2	5	2
29–42 d						
Mean	55	70	66	33	39	39
SE	3	5	2	2	3	1
43–56 d						
Mean	40	38	41	20	18	17
SE	3	4	2	1	2	3
Mean total wt gain over 8 weeks (g/rat)						
Mean	203	212	242	134	128	149
SE	6	13	3	5	3	7
Wt gain (g/g food eaten over 8 weeks)	0·29	0·28	0·29	0·22	0·21	0·21
Energy value of diets (kJ/g)	18·2	18·1	19·1	18·2	18·1	19·1
Wt gain/energy value of food eaten over 8 weeks (g/kJ)	0·016	0·015	0·015	0·012	0·012	0·011

* For details see Table 1.

Diets

The dry-powder diets tested in Expt 1 are shown in Table 1. The lactitol monohydrate, xylitol and sucrose were all in the form of coarse powders of uniform particle size.

The biscuits in Expt 2 contained 166 g sucrose or lactitol/kg, and they were incorporated in the diets at 660 g/kg, so that the final dietary level of lactitol in the test regimen was 110 g/kg. They were pulverized in a hammer-mill before the preparation of the diets (for details of composition, see Table 2).

The rats had free access to the diets from non-spill containers, and to non-fluoridated water (fluoride $< 0{\cdot}1$ μg/ml).

Table 4. *Expt 1. Whole-body tissue analyses of rats receiving diets containing lactitol (L), xylitol (X) or sucrose (S)**

(Mean values expressed on a tissue wet-weight basis)

	Males			Females		
Dietary regimen...	L	X	S	L	X	S
Moisture content (g/kg)	702	699	656	688	689	661
Fat content (g/kg)	69	73	105	71	64	105
Protein content (g/kg)	191	197	202	199	202	193
Energy value (kJ/g)	7·36	7·79	8·70	7·19	7·83	8·72
Percentage of energy retained from food	11·74	11·94	13·16	8·60	9·05	9·38
Wt of fat deposited (mg/kJ food eaten)	1·10	1·12	1·59	0·85	0·74	1·13

* For details, see Table 1.

Table 5. *Expt 1. Dental plaque and caries values for rats receiving diets containing lactitol, xylitol or sucrose†*

(Mean values with their standard errors: values for males and females combined)

Dietary regimen...	Lactitol		Xylitol		Sucrose	
	Mean	SE	Mean	SE	Mean	SE
Plaque score	11·3	0·8	7·2	0·6	12·2	0·6
Inter-group differences; statistical significance of comparison with: Lactitol group			**		NS	
Xylitol group					***	
Caries score	1·9	0·4	1·7	0·2	7·5	1·2
Inter-group differences; statistical significance of comparison with: Lactitol group			NS		***	
Xylitol group					**	
Total no. of lesions	1·9	0·3	1·7	0·2	5·5	0·6
Inter-group differences; statistical significance of comparison with: Lactitol group			NS		***	
Xylitol group					***	
Average score per lesion	1·01	0·01	1·00	0·0	1·26	0·06
Inter-group differences; statistical significance of comparison with: Lactitol group			NS		***	
Xylitol group					***	

NS, not significant. $^{**}P < 0·01$, $^{***}P < 0·001$. † For details, see Table 1.

RESULTS

Expt 1. Comparison of lactitol, xylitol and sucrose at 160 g/kg in the diet

The animals (fourteen males and eight females on each regimen) took 2–3 weeks to adapt to the lactitol diet, showing decreased food intake and increased water intake compared

Table 6. *Expt 2. Water and food intake values, and weight gains of rats receiving biscuits containing 166 g lactitol or sucrose/kg*†

	Lactitol biscuits		Sucrose biscuits	
Mean water intake (ml/rat per d)				
0–14 d	14·3		12·3	
15–28 d	19·6		19·7	
29–42 d	20·1		20·6	
43–56 d	36·5		29·0	
Mean food intake (g/rat per d)				
0–14 d	10·9		11·6	
15–28 d	16·1		14·4	
29–42 d	19·0		17·8	
43–56 d	23·1		23·3	
Wt gain (g/rat)	Mean	SE	Mean	SE
at start	40	2	42NS	2
0–14 d	58	3	72**	3
0–28 d	103	4	123***	4
0–42 d	143	5	158**	4
0–56 d	175	4	191*	5
Mean wt gain (g/g food eaten)				
0–14 d	0·123		0·187	
15–28 d	0·199		0·257	
29–42 d	0·149		0·136	
43–56 d	0·100		0·100	
Energy value of diets (kJ/g)	21·53		20·60	
Mean wt gain/energy value of food eaten over 56 d (g/kJ)	0·0065		0·0077	

NS, not significant. *$P < 0·05$, **$P < 0·01$, ***$P < 0·001$. † For details, see p. 19.

with the sucrose control during that time (Table 3). They were slower to adapt to the xylitol regimen, with an increased water intake right up to 6 weeks (Table 3). Because of their poor physical condition only eleven out of the twenty-two rats in the xylitol group were allowed to remain on the experiment for the full 8 weeks. The values for weight gains in Table 3 show that the animals on the sucrose regimen were significantly heavier than those on the two polyols, with $P < 0·01$ for the comparison of sucrose versus either polyol in both the males and females. The lactitol group gained weight significantly better than the xylitol group in the females ($P < 0·05$).

The results of the carcass analyses are given in Table 4. The main features were the higher fat content, total energy value, proportion of energy retained from the food, and weight of fat deposited per energy value of the food, on the sucrose control regimen compared with the two polyol regimens.

The dental results are given in Table 5. Significantly less plaque was recorded on the xylitol than on the other two regimens, but the plaque on the lactitol regimen was more translucent and gel-like than the normal plaque on the sucrose regimen. The differences in caries values (caries scores, total number of lesions and severity of the lesions) between the sucrose and the polyol regimens were all highly significant. The polyol regimens both produced very low levels of caries, not significantly apart. The pattern of attack was normal, with most of the lesions developing in the molar fissures, and the highest incidence in the central fissure of the first molar and the anterior fissure of the second molar.

Table 7. *Expt 2. Dental plaque and caries values of rats receiving biscuits containing 166 g lactitol or sucrose/kg*
(Mean values with their standard errors)

	Lactitol biscuits		Sucrose biscuits	
	Mean	SE	Mean	SE
Plaque score	4·2**	0·6	6·5	0·6
Caries score	7·8***	1·4	31·4	2·4
Total no. of carious lesions per rat	4·9***	0·6	12·9	0·6
Average no. of gross cavities per rat	0·9***	0·3	4·7	0·5
Average score per lesion	1·43***	0·11	2·38	0·10

$^{**}P < 0.01$, $^{***}P < 0.001$.

Expt 2. Trial of lactitol biscuits

The lactitol biscuits group showed signs of osmotic disturbance of water intake in the first 2 weeks of the experiment, with slightly increased water intake (Table 6; twenty-one female rats per group). Weight gains were consistently lower on the lactitol than on the sucrose regimen. Food conversion values (average weight gain per unit weight of food eaten) on the lactitol compared with the sucrose regimen were 34% lower in the first 2 weeks and 22% lower in the next 2 weeks of the trial, but they recovered after that.

Dental plaque scores were significantly lower on the lactitol than on the sucrose biscuits regimen (Table 7). The four separate sets of measurements recording caries attack all showed highly significant differences on replacing the sucrose in the biscuits by lactitol (Table 7). The caries score averaged 75% lower, the total number of lesions 62% lower, and the score per lesion 40% lower. In addition the average number of cavities was below 1 per animal, compared with 4·7 on the sucrose biscuits regimen.

DISCUSSION

The main finding was the low cariogenicity of lactitol replacing sucrose in the diet of the rats, confirming the observations of van der Hoeven (1986), and extending them by using lower, more realistic levels of lactitol, continuing the experiments for a longer period to allow more advanced lesions to develop, and testing lactitol in the form of a manufactured food product, sweet biscuits, rather than just as a raw food ingredient.

The general levels of caries were exceptionally high in Expt 2 because biscuits are very cariogenic in this model system, but the dental findings from both experiments are unambiguous, and cannot be explained by differences in food intake (for example, see Table 6), even though a measured-portion feeding machine was not used. They show (Expt 1) that lactitol had a cariogenic potential as low as that of xylitol, which was the best of a range of bulk sweeteners reviewed by Havenaar (1987).

A direct comparison of lactitol and xylitol is therefore of value. Van der Hoeven (1986) recorded fewer fissure lesions on a xylitol regimen than on a lactitol regimen, but it is not clear if the difference was significant. The findings reported here show very closely similar caries values on the lactitol and xylitol regimens, but the plaque scores on xylitol were lower than those on lactitol (Table 5). This raises the question of the suitability of the polyols as substrates for acid and polysaccharide production by oral micro-organisms, a key factor in determining their cariogenic potential. There is ample evidence that xylitol is not easily fermentable by oral bacteria (Linke, 1986, 1987), but relatively little information on lactitol. Imfeld (1983) reported no harmful drop in interdental plaque pH in a single test of lactitol. The results of Havenaar *et al.* (1978) suggested that although the fermentation of lactitol was slow initially, *Streptococcus mutans* could adapt to metabolize it on subculturing in media containing lactitol. More recent work showed that very little polysaccharide and acid (with consequent low enamel demineralization) were formed from lactitol by mixed cultures of plaque micro-organisms, and that comparing six different bulk sweeteners, only xylitol was less readily metabolized by the micro-organisms than lactitol (Grenby *et al.* 1989).

The indications from this limited amount of work on lactitol are therefore that it has good dental properties compared with sucrose, but its nutritional properties and metabolic effects in the mammalian body also need to be taken into account. Both of these rat experiments showed a raised water intake compared with the sucrose control in the first 2 weeks on the lactitol regimens (Tables 3 and 6), but the disturbance of water balance was much more severe and longer-lasting with xylitol at the same level (Table 3). This is a well-known property of dietary polyols, which are not absorbed from the gastrointestinal tract by any active mechanism, and attract water into the gut lumen by osmosis.

These physiological considerations are discussed by Ziesenitz & Siebert (1987), along with what is known of the pathways of metabolism of lactitol. They have not been investigated in detail, but gastrointestinal reactions were noted in fifteen out of sixteen people eating sweets made with lactitol instead of sucrose, although plaque deposition on their teeth was significantly diminished (Grenby & Desai, 1988).

One use for lactitol put forward by den Uyl (1987) is in reduced-energy foods. Ziesenitz & Siebert (1987) observed that people receiving 50 g lactitol/d for 4 d showed 50% less energy utilization than with sucrose. In rats, too, in the first of the two trials reported here (Table 4), comparing the lactitol with the sucrose control regimen, fat storage was 33% lower, and the mean total energy stored was 34–35% lower.

These factors have to be taken into consideration in the formulation of foods containing lactitol. A range of food products has been prepared (den Uyl, 1987), but only two of them have been tested for their dental properties: sweets in a trial in man (Grenby & Desai, 1988) and biscuits in the second of the experiments in rats reported here. Both gave favourable findings, with a difference in the mean caries scores of no less than 75% on replacing the sucrose in conventional biscuits by lactitol at a final dietary content of 110 g/kg (Table 7). Further work is now required on other types of food, and to establish optimum dietary levels of lactitol replacing sugars.

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