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Genetic lactase non-persistence, consumption of milk products and intakes of milk nutrients in Finns from childhood to young adulthood

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Previous evidence suggests that the lactase gene C/T_{-13910} polymorphism (rs4988235) is associated with avoidance of milk products and lower Ca intake. We examined whether the consumption of milk and milk products and the intakes of milk nutrients differ between the lactase genotypes from childhood to young adulthood. Subjects belong to the Cardiovascular Risk in Young Finns Study where the first cross-sectional surveys were conducted in 1980 (*n* 3596), with follow-up studies in 1983, 1986, 1989, 1992 and 2001 (*n* 2620). The same dietary questionnaire was used throughout the follow-up to collect data on habitual consumption of milk and milk products in all subjects, and daily nutrient intakes were assessed with 48 h dietary recalls in 50 % of the subjects. Subjects with the lactase non-persistence (C/C_{-13910}) genotype consumed less milk since childhood, but the consumption of other milk products did not differ between the genotypes. In adult females, the lactose content of milk products consumed was lower ($P=0{\cdot}003$), and in both sexes low-lactose and milk-free diets were more common in the C/C_{-13910} genotype than in the other genotypes. Inadequate Ca intake was most common in females with the C/C_{-13910} genotype as early as in childhood (15–63 %), but in males only in adulthood (24 %). In adult females, preference for low-lactose milk and milk products equalised the differences in Ca intake between the genotypes. Thus, in those with the C/C_{-13910} genotype, preference for low-lactose milk and milk products may decrease the risk for inadequate Ca intake.

Lactase genotype: Milk consumption: Calcium intake: Lactose intake

More than half of the world's population have a deficit in the enzyme lactase (lactase non-persistence)[1]. Lactase non-persistence results in the presence of maldigested lactose in the colon, which may cause unpleasant symptoms such as bloating, abdominal pain and diarrhoea. The most common therapeutic approach to lactase non-persistence is to exclude milk and milk products from the diet[2], even though this may predispose to poorer bone health due to lowered Ca intake[3–6]. However, the evidence is not consistent[7,8], and the effectiveness of milk and milk product consumption in the prevention of osteoporosis has recently been re-evaluated[9].

Genetic lactase non-persistence is a recessively inherited condition caused by a decline in the activity of lactase-phlorizin hydrolase in the small intestine during maturation. Enattah *et al.*[10] found that lactase persistence correlates strongly with the C/T_{-13910} polymorphism located 13·9 kb upstream from the lactase gene. *In vitro* studies suggest that this polymorphism enhances lactase gene expression[11,12] by regulating the binding of transcription factors such as Oct-1[13]. The expression of the lactase gene in the intestinal mucosa is several times higher for the T_{-13910} allele[14]. Therefore, both the T/T_{-13910} and C/T_{-13910} genotypes possess sufficient enzyme activity to be classified as lactase persistent, and the homozygous C/C_{-13910} genotype is classified as lactase non-persistent.

Austrian and Estonian studies have shown that the C/C_{-13910} genotype is associated with a lower consumption of milk and milk products, lower Ca intake and impaired Ca absorption[15–18]. In Finland dairy products have traditionally constituted an important part of the daily diet, and milk and milk products provide about two-thirds of the Ca intake of adults in Finland[19]. Hence, genetic lactase non-persistence in Finns could be considered a potential risk factor for low Ca intake by limiting the consumption of milk and milk products. Some Finnish studies indicate that the C/C_{-13910} genotype is linked to milk avoidance and a low-lactose diet in childhood, adolescence and adulthood[20–22]. However, Finnish studies in young men[23], postmenopausal women[24]

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and elderly individuals[25] have observed no differences in the consumption of milk products or in total Ca intakes between the lactase genotypes. It is noteworthy that so far no evidence has been published on the differences in habitual lactose intake between the lactase genotypes.

We have investigated the C/T_{-13910} polymorphism in the Cardiovascular Risk in Young Finns Study, which provides a prospective 21-year follow-up and well-documented longitudinal data on food consumption and nutrient intakes from early childhood until young adulthood[26–31]. A recent report from the Young Finns Study showed that the lactase gene C/T_{-13910} polymorphism was not associated with mean growth speed but contributed significantly to milk product consumption and dietary Ca intake from childhood into young adulthood[32]. In this present analysis, we further examine whether the type of milk and milk products consumed and nutrient intakes from milk and milk products differ between the lactase genotypes from childhood into young adulthood. Since low-lactose milk and milk products are commonly used in Finland, our specific aim was to compare the lactose content of milk products consumed between the lactase genotypes.

Subjects and methods

Cardiovascular Risk in Young Finns Study

The Cardiovascular Risk in Young Finns Study is an ongoing multi-centre follow-up of atherosclerosis risk factors for young Finns. The first cross-sectional survey was conducted in 1980[26]. The total sample comprised 4320 subjects aged 3, 6, 9, 12, 15 and 18 years. The subjects were randomly selected from the national population register from five university cities in Finland (Helsinki, Turku, Tampere, Kuopio and Oulu) and the rural municipalities in their vicinity. A total of 3596 subjects (83 % of those invited) participated in 1980. The same subjects were re-examined in follow-up studies in 1983 and 1986, as well as in a 21-year follow-up in 2001 at the age of 24–39 years (Table 1)[28,30]. In 1989 and 1992, only smaller subgroups of the original cohort were studied. In order to compare the consumption of milk and milk products between the lactase genotypes at the ages of 6, 9, 12, 15 and 18 years, we combined the data from the different follow-up years as marked in Table 1. Subjects provided their written informed consent, and the local ethics committees of the participating universities approved the study protocol[26].

Dietary data

Previous studies have described the details of the dietary study included in the project[27,29,31]. The consumption of milk and milk products was assessed with a dietary questionnaire on habitual eating behaviour and food choices, assessed at baseline in 1980 and in the follow-ups in 1983, 1986, 1989, 1992 and 2001. The questionnaire has been slightly modified over the years to fit the relevant issues according to the follow-up year and subjects' age, but has included the same set of food choices during all follow-ups. The questionnaire has always included questions on the habitual amount of milk consumed (glasses per d) and the frequency of consumption of cheese, sour milk products (buttermilk, sour whole milk, yoghurt and curd cheese) and ice cream. Since the questionnaire has been designed to assess milk fat intake, the questionnaire included a question on the type of the milk products based on their fat content. Otherwise the different milk product brands were not outlined and the lactose content of the products was not assessed. On the basis of the questionnaire data, we converted the reported frequency of consumption of milk products into portions per week (p/w) as follows: 'daily' = 7 p/w; 'nearly every day' = 5 p/w; 'a couple of times weekly' = 2·5 p/w; 'once weekly' = 1 p/w; 'once or twice monthly' = 0·5 p/w; 'less or never' = 0 p/w. We then added this figure up with the number of glasses of milk reported, thus resulting in an index variable representing each subject's habitual consumption of milk and milk products as portions per week (or per d when divided by 7). If the subjects answered none to the question 'how many glasses of milk was consumed daily', they were classified as not drinking milk. In the questionnaire the subjects reported if they followed a low-lactose or milk-free diet, but this information was based on their own reporting and was not verified with the clinical diagnosis of lactose intolerance.

The data on the subjects' daily intake of energy, Ca, lactose and protein were obtained with a detailed 48 h dietary recall assessed in 50 % of subjects at baseline in 1980 and in follow-ups in 1986 and 2001. Dietary interviewers were all trained dietitians and collected information on foods and beverages consumed by subjects during the 2 d before the interview. In 1980 and 1986, 3- to 12-year-old children were interviewed together with their mothers, fathers or another accompanying person. As detailed information as possible on the type and amount of foods and drinks consumed was documented on a form by the interviewer. The food composition data in the 1980s were based on the Finnish food composition tables (maintained by the University of Helsinki) and on analytical data obtained from the

Table 1. Subjects and dietary studies in the Cardiovascular Risk in Young Finns Study

Year	Subjects (*n*)	%	Dietary data	Age cohorts*
1980	3596	100	Questionnaire and 48 h recall (50 %)	3 (6 9) (12 15) (18 21) 24 27 30
1983	2991	83	Questionnaire	(6 9) (12 15) (18 21) 24 27 30
1986	2799	78	Questionnaire and 48 h recall (50 %)	(9 12) (15 18) (21 24) 27 30
1989	350†	10	Questionnaire	(12 15) (18 21) (24 27) 30
1992	891†	25	Questionnaire	(15 18) (21 24) (27 30)
2001	2620	73	Questionnaire and 48 h recall (50 %)	(24 27) (30 33) (36 39)

* Bracketing around ages indicates the age groupings used.
† Measurements made only for subgroups.

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local food industry. Foreign food composition tables were used when no appropriate domestic data were available. After 1980 the food composition tables were passed to the responsibility of the National Public Health Institute. In 2001, the latest version of the National Food Composition Database was used to calculate the intakes of nutrients[33]. Ca intake that did not meet the Finnish nutrition recommendations was considered as inadequate (aged 6–9 years 700 mg/d; 10–18 years 900 mg/d; adults 800 mg/d)[34]. The dietary data for adulthood in 2001 provide information on the food sources of nutrients, thus enabling us to calculate the subjects' intakes of Ca, lactose, protein and vitamin D separately from different types of milk products. The food composition table used in 2001 also allowed us to calculate the lactose content of different types of cultured milk products such as buttermilk, sour whole milk, yoghurt and curd cheese. Based on the dietary recall data, the lactose content of milk and milk products consumed was calculated as g lactose per 100 g milk and milk products consumed. This calculated variable was used as an indicator of preference for low-lactose milk and milk products and was divided into tertiles by sex in order to compare Ca intake between the tertiles. In females, the tertile limits were (g/100 g milk products) $<3{\cdot}2$ for low, 3·2–4·1 for middle and $>4{\cdot}1$ for high, and in males $<3{\cdot}5$, 3·5–4·3 and $>4{\cdot}3$, respectively.

Lactase C/T$_{-13910}$ genotyping

Blood samples were collected for genetic analysis in 2001. DNA was extracted from peripheral blood leucocytes with a commercially available kit (Qiagen Inc., Hilden, Germany). Lactase C/T$_{-13910}$ genotyping (rs4988235) was performed by employing a 5′-nuclease assay, and fluorogenic allele-specific TaqMan probes and primers[35] were used with the ABI Prism 7000 Sequence Detection System (Applied Biosystems, Foster City, CA, USA). Known control samples were run in parallel with unknown DNA samples. Lactase C/T$_{-13910}$ polymorphism was measured in 2265 adults.

Statistical analysis

Statistical analyses were performed with SPSS 12.0 for Windows (SPSS Inc., Chicago, IL, USA). The Hardy–Weinberg equilibrium of the C/T$_{-13910}$ polymorphism was performed using the exact test of the Genepop software (http://wbiomed.curtin.edu.au/genepop)[36]. The homogeneity of variances across the lactase genotypes was tested with Levene's statistics. The differences in milk and milk product consumption and nutrient intakes from milk between the lactase genotypes were tested with one-way ANOVA, and for the variables with unequal variances with Kruskall–Wallis non-parametric analysis. Two-way ANOVA was used to analyse how the interaction between lactase genotype and lactose content of milk and milk products consumed was associated with daily Ca intake. With significant ANOVA *P* values, Fisher's least significant difference method was used for pairwise *post hoc* comparisons, and the Bonferroni method was applied to non-significant *P* values. If the Levene's test results were significant, we used the non-parametric Games–Howell *post hoc* test. Pearson's χ^2 was used to compare the proportions of those with inadequate Ca intake with those with recommended Ca intake, milk drinkers with non-milk drinkers, and the proportions of those following a low-lactose or milk-free diet with those following a diet with normal lactose content within the lactase genotypes. Females and males were analysed separately and also pooled when Ca intake in adulthood was analysed. A *P* value of less than 0·05 was considered significant.

Results

The distribution of lactase genotypes was in Hardy–Weinberg equilibrium ($P=0{\cdot}814$) (Table 2). Age, height, weight or energy intake in adulthood did not differ between the lactase genotypes in either sex (data not shown). When we compared milk and milk product consumption between the lactase genotypes, the most profound differences at all age points in both sexes were found in the consumption of milk (Table 2). Cheese consumption tended to be higher for the C/C$_{-13910}$ genotype, but the genotypes differed significantly only in 6-year-old males. The consumption of sour milk products did not differ between the lactase genotypes in either sex or at any age. The consumption of ice cream was so low that it was not included in the analyses.

In females, there was a trend of lower daily Ca and lactose intakes for the C/C$_{-13910}$ genotype since 9 years of age, although only Ca intake at the age of 9 years differed significantly between the genotypes (ANOVA $P=0{\cdot}025$, C/C *v.* T/T $P=0{\cdot}06$ and C/C *v.* C/T $P=0{\cdot}007$) (Fig. 1). In males, a similar non-significant trend of difference in Ca and lactose intakes between the genotypes was found only at the age of 18 years. In adulthood, the C/C$_{-13910}$ genotype was associated with a lower daily Ca intake in both sexes (Table 3). However, the differences between the genotypes were significant only when the sexes were pooled (T/T = 1271 mg, C/T = 1322 mg and C/C = 1167 mg; ANOVA $P=0{\cdot}009$, C/C *v.* T/T $P=0{\cdot}053$ and C/C *v.* C/T $P=0{\cdot}002$). In adulthood, daily lactose intake was lowest for the C/C$_{-13910}$ genotype in both sexes (Games–Howell: C/C *v.* T/T $P<0{\cdot}001$ for females and $P=0{\cdot}003$ for males, C/C *v.* C/T $P<0{\cdot}001$ for both sexes). Adult females with the C/C$_{-13910}$ genotype had the lowest protein intake (Fisher's least significant difference: C/C *v.* T/T $P=0{\cdot}37$ and C/C *v.* C/T $P=0{\cdot}03$). The daily intake of vitamin D in adulthood did not differ between the genotypes.

In adulthood, milk and milk products provided 72 % of total Ca intake for the T/T and C/T$_{-13910}$ genotypes and 68 % for the C/C$_{-13910}$ genotype in females, and 72, 76 and 70 % for the genotypes T/T, C/T and C/C in males, respectively (for values, see Table 3). The intakes of Ca, lactose, protein and vitamin D from milk were significantly lowest for the C/C$_{-13910}$ genotype in both sexes, but the intakes of these nutrients from cheese or sour milk products did not differ between the genotypes. In addition, the lactose content of milk and milk products consumed was lowest for those with the C/C$_{-13910}$ genotype; however, the difference was significant only in females (Fisher's least significant difference: C/C *v.* T/T $P=0{\cdot}03$ and C/C *v.* C/T $P=0{\cdot}001$).

In females, the proportions of those with inadequate Ca intake were highest in the C/C$_{-13910}$ genotype at all ages (15–63 %); however, the differences were significant only at the ages of 9 and 15 years and in adulthood (Table 4).

Table 2. Consumption of milk and milk products (dietary questionnaire) by age and lactase genotype
(Mean values and standard deviations)

Lactase genotype…		Females								Males						
		T/T		C/T		C/C				T/T		C/T		C/C		
	Subjects (*n*)	Mean	SD	Mean	SD	Mean	SD	*P**	Subjects (*n*)	Mean	SD	Mean	SD	Mean	SD	*P**
Genotype distribution (*n*)		425		616		215				339		500		186		
Genotype distribution (%)		34		49		17				33		49		18		
Milk index (portions/d)†																
6 years	317	4·2	1·5	4·7	1·4	3·7	1·4	<0·001	244	4·9	1·8	4·5	1·6	4·5	1·7	0·31
9 years	529	4·3	1·6	4·5	1·6	4·0	1·5	0·07	426	5·2	1·8	4·5	1·7	4·7	1·6	0·001
12 years	702	4·5	1·7	4·6	1·8	3·9	1·9	0·002	564	5·2	1·9	4·9	2·0	4·7	2·3	0·17
15 years	767	4·1	2·0	3·9	1·7	3·8	1·9	0·37	592	5·3	2·1	5·2	2·2	4·6	2·1	0·012
18 years	710	3·3	1·6	3·2	1·5	2·9	1·6	0·12	558	5·3	2·3	4·9	2·3	4·8	2·0	0·08
Adult (24–39 years)	1256	2·7	1·7	2·7	1·6	2·2	1·3	<0·001‡	1025	3·5	2·1	3·4	2·1	2·6	1·7	<0·001‡
Milk (glasses/d)																
6 years	317	3·0	1·4	3·4	1·4	2·7	1·4	0·001	244	3·7	1·7	3·3	1·5	3·3	1·7	0·2
9 years	529	3·1	1·6	3·3	1·6	2·9	1·5	0·14	426	4·0	1·7	3·4	1·7	3·4	1·6	0·001
12 years	702	3·3	1·7	3·4	1·8	2·7	1·8	0·001	564	4·0	1·8	3·8	2·0	3·5	2·1	0·02‡
15 years	767	3·0	1·9	2·9	1·7	2·6	2·0	0·11	592	4·2	2·1	4·0	2·1	3·5	2·1	0·01
18 years	710	2·4	1·7	2·2	1·6	1·8	1·5	0·001	558	4·2	2·3	4·0	2·4	3·7	2·2	0·17
Adult (24–39 years)	1256	1·4	1·6	1·5	1·5	0·8	1·3	<0·001‡	1025	2·3	2·0	2·2	2·1	1·5	1·7	<0·001‡
Cheese (portions/week)																
6 years	317	3·6	2·5	3·8	2·4	3·1	2·3	0·14	244	3·2	2·6	3·8	2·6	4·5	2·4	0·02
9 years	529	3·8	2·6	4·0	2·6	4·0	2·4	0·76	426	3·6	2·8	3·8	2·6	3·9	2·9	0·55
12 years	702	4·0	2·5	4·3	2·5	4·5	2·4	0·15	564	3·8	2·8	3·8	2·6	4·4	2·6	0·1
15 years	767	3·9	2·4	3·8	2·4	3·6	2·4	0·4	592	3·8	2·7	3·9	2·4	3·8	2·7	0·87‡
18 years	710	3·9	2·5	4·1	2·4	4·2	2·5	0·62	558	4·0	2·6	3·9	2·7	3·9	2·5	0·99
Adult (24–39 years)	1256	4·9	2·8	4·8	2·8	5·2	2·7	0·17	1025	4·5	2·9	4·8	2·8	4·9	2·4	0·11
Sour milk products (portions/week)																
6 years	317	3·7	2·4	3·8	2·6	3·2	2·5	0·27	244	3·6	2·5	3·8	2·5	3·4	2·4	0·52
9 years	529	3·7	2·7	3·7	2·6	3·2	2·8	0·29	426	3·6	2·6	3·4	2·5	3·8	2·9	0·60‡
12 years	702	3·4	2·5	3·3	2·6	3·2	2·5	0·77	564	3·5	2·7	3·4	2·6	3·4	2·6	0·98
15 years	767	2·8	2·4	2·7	2·4	2·6	2·4	0·8	592	3·0	2·6	2·8	2·4	3·0	2·6	0·76
18 years	710	2·8	2·5	2·9	2·6	2·7	2·5	0·66	558	3·1	2·6	2·7	2·6	2·5	2·4	0·14
Adult (24–39 years)	1256	3·5	2·4	3·4	2·5	3·4	2·4	0·8	1025	2·8	2·4	2·9	2·5	2·5	2·4	0·08‡

* One-way ANOVA.
† Reported frequency of consumption of milk and milk products (portions/d).
‡ Kruskall–Wallis non-parametric test.

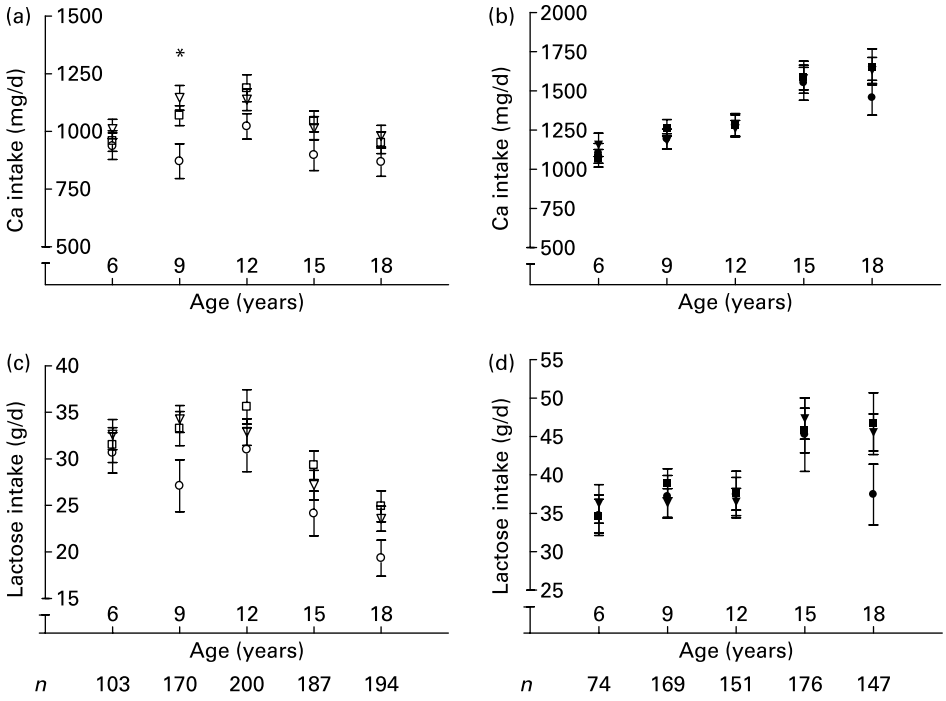

Fig. 1. Intakes of (a) Ca in females (mg/d), (b) Ca in males, (c) lactose in females (g/d) and (d) lactose in males in relation to age by the lactase genotypes: in females T/T (□); C/T (▽); C/C (○) and in males T/T (■); C/T (▼); C/C (●). Lactose and Ca intakes were assessed with a detailed 48 h dietary recall. Values are means, with standard errors represented by vertical bars. * Ca intake in 9-year-old females differed between the lactase genotypes: ANOVA $P = 0{\cdot}025$, C/C *v.* T/T $P = 0{\cdot}06$ and C/C *v.* C/T $P = 0{\cdot}007$.

In males, inadequate Ca intake was more common in the C/C_{-13910} genotype (24 %) than in other genotypes only in adulthood. The proportions of those who reported not drinking milk were highest in adulthood, being over half in females and over one-third in males with the C/C_{-13910} genotype. In adulthood, those with the C/C_{-13910} genotype reported following low-lactose or milk-free diets more often than did those with C/T or T/T_{-13910} genotypes. In both sexes, the avoidance of lactose and milk increased with age in all lactase genotypes.

In both sexes, those in the lowest lactose content tertile consumed more cheese and sour milk products and less milk than those in the second and third tertiles (Table 5). Adult females with the C/C_{-13910} genotype who consumed milk and milk products with higher lactose content had lower Ca intake than those in other genotypes (genotype × lactose content of milk products consumed $P = 0{\cdot}022$) (Fig. 2). In lower lactose content tertiles, Ca intake did not differ between the lactase genotypes. In males, the trend was similar but not significant (genotype × lactose content of milk products consumed $P = 0{\cdot}136$).

Discussion

The primary aim of the present study was to investigate the differences in milk and milk product consumption and nutrient intakes from milk and milk products between the lactase genotypes, since previous evidence is neither comprehensive nor consistent[15–17,20–25]. Furthermore, this is the first study to examine the differences in the type of milk products consumed and in the lactose intake between the lactase genotypes in a follow-up setting from childhood into adulthood.

We showed that already in childhood, those with the C/C_{-13910} genotype consumed less milk than did those with the other genotypes, and the proportions of those not drinking milk were greatest in the C/C_{-13910} genotype as early as in childhood. These results are in congruence with earlier findings from Finnish studies[20,21]. In addition, those with the C/C_{-13910} genotype tended to eat more cheese than did those with other genotypes, although the differences were not significant and not distinct at all age points. We found no differences in the habitual consumption of sour milk products between the lactase genotypes, although lactose maldigesters do tolerate fermented milk products better than non-fermented[37]. It is noteworthy that until the age of 12 years, the consumption of milk and milk products in males with the C/C and C/T_{-13910} genotypes was very similar and lower than for the T/T_{-13910} genotype. In females with the C/C_{-13910} genotype milk and milk product consumption was lowest since the age of 6 years. These findings could result from sex differences in the manifestation age of genetic lactase non-persistence or in the magnitude of the T_{-13910} variant expression, but evidence is lacking. One possible explanation could be that females and males differ in their sensitivity to gastrointestinal symptoms caused by maldigested lactose. Previous studies have shown that women with lactose maldigestion may experience stronger symptoms than men with lactose maldigestion[38,39]. The present results support these findings because low-lactose or milk-free diets were almost twice as common in females with the C/C_{-13910} genotype than in males with the same genotype.

A glass of milk or sour milk is a typical meal component in Finland both for children and adults. In addition, milk products are commonly used as components of traditional Finnish

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Table 3. Dietary intakes of selected nutrients (48 h dietary recall) in adulthood (24–39 years) by lactase genotype
(Mean values and standard deviations)

Lactase genotype…	Females							Males						
	T/T		C/T		C/C			T/T		C/T		C/C		
	Mean	SD	Mean	SD	Mean	SD	*P**	Mean	SD	Mean	SD	Mean	SD	*P**
Subjects (*n*)	193		273		100			152		232		81		
Subjects (%)	34		48		18			33		50		17		
Ca intake (mg/d)														
All sources	1160	470	1174	479	1048	462	0·07	1413	727	1497	661	1313	563	0·09
Milk products as sources of Ca	886	468	889	466	766	447	0·06	1100	696	1183	634	992	537	0·06
Milk	332	321	361	300	215	229	<0·001†	523	467	553	517	351	305	0·009†
Cheese	382	306	371	322	376	326	0·93	438	412	476	376	524	397	0·28
Sour milk products	143	156	128	162	149	178	0·44	111	176	124	220	92	141	0·41
Lactose intake (g/d)														
All sources	18·8	13·2	19·3	13·3	13·1	9·8	<0·001†	26	19·9	28·1	21·5	18·7	13·1	0·003†
Milk products as sources of lactose	17·6	13·2	18·3	13·1	12	9·5	<0·001†	24·7	19·8	26·5	21·3	17·3	12·7	0·003†
Milk	13·3	23·2	14·4	12·6	8	8·92	<0·001†	21·3	19·1	22·7	21·2	14·4	13	0·006†
Cheese	0·2	0·44	0·19	0·54	0·17	0·38	0·86	0·2	0·49	0·12	0·31	0·14	0·38	0·13
Sour milk products	3·15	3·87	2·65	3·71	2·91	3·95	0·38	2·13	3·33	2·65	5·21	1·86	2·9	0·29
Lactose content of milk products (g/100 g)‡	3·44	1·09	3·6	1·03	3·16	1·15	0·003	3·72	1·03	3·74	1·06	3·44	1·14	0·09
Protein intake (g/d)														
All sources	68·9	19·8	68·9	27·1	66·3	19·2	0·05	96	35	102·9	34·3	98·3	26·1	0·13
Milk products as sources of protein	24·7	13·2	24·9	12·9	21·2	12	0·012	30·6	19	32·5	17·4	28·2	15·7	0·15
Milk	8·69	8·39	9·42	7·79	5·59	5·92	<0·001†	13·7	12·1	14·5	13·5	9·2	8·03	0·010†
Cheese	11·7	9·54	11·5	9·87	11·3	9·06	0·94	13·4	11·8	14·2	10·9	15·9	12·3	0·28
Sour milk products	3·6	3·83	3·24	3·72	3·72	4·47	0·47	2·78	4·46	3·15	5·58	2·38	3·62	0·45
Vitamin D intake (μg/d)														
All sources	3·69	3·48	4·94	19·3	3·3	2·58	0·47	5·61	6·91	4·76	4·49	4·2	3·15	0·86†
Milk products as sources of vitamin D	0·35	0·23	0·38	0·24	0·3	0·2	0·043	0·49	0·38	0·51	0·39	0·34	0·23	0·005†
Milk	0·19	0·21	0·21	0·22	0·13	0·17	<0·001†	0·31	0·35	0·32	0·38	0·17	0·18	0·003†
Cheese	0·09	0·08	0·09	0·08	0·09	0·08	0·9	0·11	0·1	0·11	0·09	0·13	0·1	0·29
Sour milk products	0·05	0·07	0·05	0·09	0·06	0·09	0·59	0·04	0·1	0·05	0·11	0·02	0·05	0·2

* One-way ANOVA.
† Kruskall–Wallis non-parametric test.
‡ The lactose content of milk products was calculated as g lactose per 100 g milk and milk products consumed.

Table 4. The numbers and proportions (%) of those who have inadequate Ca intake, who do not drink milk or who report following a low-lactose or milk-free diet by lactase genotype

	Females							Males						
	T/T		C/T		C/C		χ^2 test	T/T		C/T		C/C		χ^2 test
	n/*N*	%	*n*/*N*	%	*n*/*N*	%	*P*	*n*/*N*	%	*n*/*N*	%	*n*/*N*	%	*P*
Ca intake lower than recommended*														
6 years	4/33	12	7/50	14	3/20	15	0·95	3/23	13	5/35	14	1/16	6	0·71
9 years	11/62	18	17/85	20	10/23	44	0·03	4/63	6	12/80	5	2/26	8	0·22
12 years	15/60	25	36/107	34	9/33	27	0·47	8/48	17	13/67	19	10/36	28	0·44
15 years	28/71	39	34/86	40	19/30	63	0·05	10/59	17	7/89	8	3/28	11	0·23
18 years	28/61	46	48/96	50	22/36	61	0·34	6/46	13	12/78	15	2/23	9	0·71
Adult (24–39 years)	44/193	23	58/273	21	36/100	36	0·01	27/152	18	23/231	10	19/81	24	0·006
Does not drink milk†														
6 years	7/115	6	2/167	1	6/56	11	0·006	1/90	1	7/129	5	5/44	11	0·03
9 years	12/194	6	26/273	6	8/78	10	0·37	5/151	3	21/191	10	3/85	3	0·02
12 years	19/251	8	23/367	6	20/116	17	0·001	11/196	6	22/296	7	10/115	9	0·56
15 years	39/321	12	49/456	11	26/155	17	0·14	12/252	5	24/363	7	13/138	9	0·20
18 years	52/299	17	78/449	17	37/153	24	0·14	15/235	6	22/333	7	10/117	9	0·73
Adult (24–39 years)	168/425	40	219/616	36	118/215	55	<0·001	80/339	24	144/500	29	71/186	38	0·002
Low-lactose or milk-free diet‡														
6 years	0/47	0	0/70	0	0/30	0	–	0/71	0	0/106	0	0/37	0	–
9 years	0/123	0	0/184	0	0/39	0	–	0/118	0	2/158	1	1/70	1	0·45
12 years	0/167	0	2/247	1	1/85	1	0·44	0/171	0	0/248	0	1/93	1	0·11
15 years	0/214	0	2/301	1	1/104	1	0·42	0/227	0	3/316	0·9	2/123	2	0·21
18 years	0/103	0	3/147	2	1/58	2	0·36	0/102	0	0/155	0	1/55	2	0·45
Adult (24–39 years)	20/309	7	29/453	6	34/152	22	<0·001	15/276	5	24/420	6	21/171	12	0·009

* Finnish Nutrition Recommendations [34] for Ca intake: children 6–9 years, 700 mg/d; children and adolescents of 10–20 years, 900 mg/d; adults, 800 mg/d.
† The questionnaire of dietary habits included a question on how many glasses of milk the subjects drink daily, and those who reported none were classified as not drinking milk.
‡ The questionnaire of dietary habits included a question on whether subjects followed a diet with low lactose content or a milk-free diet.

dishes such as soups, porridges and desserts. In Finland, unlike in most European countries, low-lactose milk and milk products have been on the market and in common use since the 1980s. Wide selections of low-lactose milks, sour milks, yoghurts and other milk products are available in most grocery stores at affordable prices. Because many Finns are aware of lactose non-persistence and familiar with low-lactose milk and milk products, these products are also widely used in institutional kitchens, restaurants, cafés and by the food industry. Therefore, it was expected that those with the C/C_{-13910} genotype followed low-lactose or milk-free diets more often than those with lactase persistence genotypes. The avoidance of milk and lactose in the diet was more common in the C/C_{-13910} genotype in adulthood than in childhood and adolescence, although previous findings from Finnish studies suggest that in the majority of children aged 8 years and in all children older than 12 years, the C/C_{-13910} genotype was already associated with very low lactase activity (< 10 U/g protein) and lower milk consumption[20]. Furthermore, the lactose content of milk and milk products chosen was lowest in females with the C/C_{-13910} genotype and in males the difference between genotypes was borderline significant. Nevertheless, the proportion of those not drinking milk and those who followed a low-lactose diet increased with age in all lactase genotypes in both sexes, which is in congruence with other studies showing a poor correlation between genetically confirmed lactase non-persistence and self-reported lactose intolerance[16,23]. However, in Finland, the consumption of milk has decreased over the past decades, and simultaneously the consumption of other milk products has increased[19], which explains why the proportion of milk drinkers decreased among all genotypes. In adulthood, the proportions of those not drinking milk were higher than proportions of those following low-lactose or milk-free diets. This finding could be

Table 5. Milk product consumption in the tertiles of lactose content in milk products consumed
(Mean values and standard deviations)

	Female (*n* 544)						Male (*n* 443)					
	1st < 3·2		2nd 3·2–4·1		3rd > 4·1		1st < 3·5		2nd 3·5–4·3		3rd > 4·3	
Tertiles of lactose content in milk products consumed (g/100 g)	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Cheese (portions/d)***	0·77	0·39	0·73	0·38	0·61	0·39	0·80	0·35	0·64	0·41	0·62	0·39
Sour milk products (portions/d)***	0·55	0·36	0·52	0·33	0·41	0·34	0·46	0·36	0·45	0·36	0·29	0·29
Milk (portions/d)***	0·43	0·90	1·36	1·28	2·27	1·76	0·76	1·12	2·19	1·66	3·41	2·05

***P<0·001 for both sexes (ANOVA).

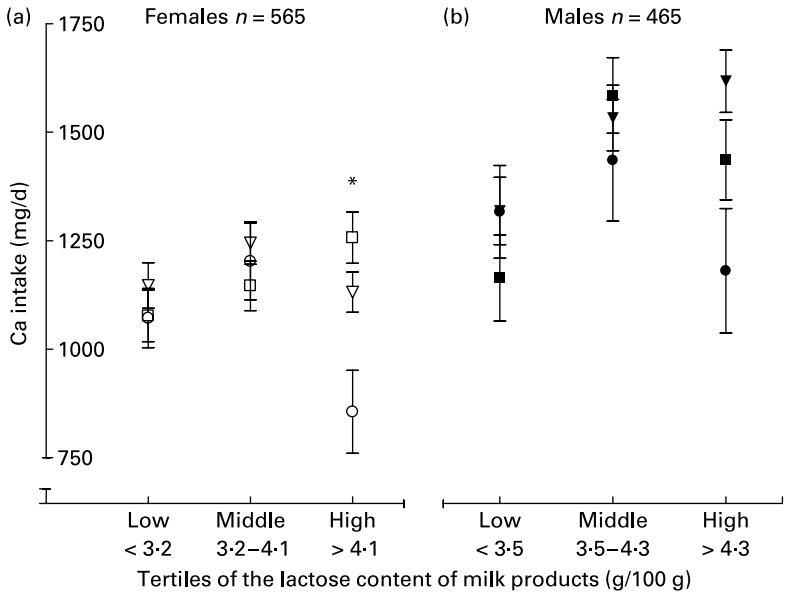

Fig. 2. (a) Ca intake in tertiles of the lactose content of milk and milk products consumed in adult females (*n* 565) (age range 24–39 years) by lactase genotypes: T/T (□); C/T (▽); C/C (○). (b) Ca intake in tertiles of the lactose content of milk and milk products consumed in adult males (*n* 465) (age range 24–39 years) by lactase genotypes: T/T (■); C/T (▼); C/C (●). Data on Ca intake and milk consumption were assessed by 48 h dietary recall. The lactose content of milk products was calculated as g lactose per 100 g milk and milk products consumed. Values are means, with standard errors represented by vertical bars. * In females, two-way ANOVA for lactase genotype $P = 0{\cdot}064$ and for interaction genotype × lactose content of milk products consumed $P = 0{\cdot}022$. In males, two-way ANOVA for lactase genotype $P = 0{\cdot}092$ and for interaction genotype × lactose content of milk products consumed $P = 0{\cdot}136$.

explained by the result suggesting that those who get symptoms from lactose or consumption of milk and milk products give up milk drinking and consume regularly other milk products that contain less lactose such as hard cheese, which is the most commonly used cheese type in Finland[19], and sour milk products. Thus, only some lactose intolerants choose low-lactose milk and milk products where lactose has been enzymically digested.

In childhood and adolescence, the average daily lactose intake in the C/C_{-13910} genotype varied from 19 to 36 g in females and from 35 to 46 g in males, and was 13 g and 19 g for adult females and males, respectively. Previous studies on the C/T_{-13910} polymorphism have reported no habitual lactose intakes; thus we could not compare the lactose intake of the C/C_{-13910} genotype in different populations. Considering the previous evidence on lactose intake in those with non-genetically confirmed lactose maldigestion[40–42], especially in childhood the lactose intakes of our subjects with the C/C_{-13910} genotype were quite high. Vesa *et al.* [40] have shown that lactose maldigesters reported no differences in the severity of symptoms after consuming milk without lactose or milk with varying amounts of lactose (0·5–7 g per 200 ml). In addition, Hertzler *et al.* [41] have shown that gastrointestinal symptoms from lactose maldigestion, measured with the hydrogen breath test, increase with doses exceeding 12 g lactose. A recent meta-analysis confirmed that the severity of gastrointestinal problems reported by lactose maldigesters did not differ after a lactose dose of 12 g (about 240 ml milk) or after ingesting a placebo under masked conditions[42]. However, individual differences in lactose tolerance may be significant, since the development of intolerance symptoms depends not only on the dose of lactose, but also on the rate of gastric emptying and delivery of lactose to the colon[43,44], and the adaptation of colonic bacteria to frequent lactose consumption[45]. Furthermore, studies have shown that maldigesters who have experienced gastrointestinal problems after the consumption of large amounts of lactose may be psychologically sensitised to milk consumption[46]. Lactose is known to facilitate Ca absorption, but previous evidence suggests that at normal intake levels lactose does not seem to significantly affect the absorption of Ca from milk in lactase-deficient or lactase-sufficient subjects[47–49]. However, a recent study by Obermayer-Pietsch *et al.* [18] showed that genetic lactase non-persistence is associated with decreased fractional Ca absorption when Ca is ingested with high amounts of lactose. Thus, together with our findings, these results suggest that a diet with lower lactose content may have beneficial effects on Ca intake and bioavailability in those with genetic lactase non-persistence.

Our previous results from the Cardiovascular Risk in Young Finns study showed that the lactase genotypes contributed significantly to dietary Ca intake from childhood into young adulthood[32]. Because sufficient Ca intake is required for the optimal attainment of peak bone mass[50,51], adequate Ca intake in children and adolescents should be assured. However, the differences in Ca intake between the lactase genotypes were less distinct than in other populations[15–17], and the habitual Ca intakes of adult Finns are higher than those of adults in other European countries[19,52]. Nevertheless, in one-third of adult females and in one-quarter of males with the C/C_{-13910} genotype, the daily Ca intake failed to meet the Finnish nutrition recommendations[34]. The lower total Ca intake in the C/C_{-13910} genotype was mostly explained by lower milk consumption, and the higher cheese consumption only partly compensated for the genotype difference in daily Ca intake. It is noteworthy that those with

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the C/C$_{-13910}$ genotype had equal Ca intake when compared with other genotypes if they chose milk and milk products with lower lactose content.

The data on milk product consumption were collected by the semi-quantitative frequency questionnaire that was used to assess dietary habits on a long-term basis. The major limitation in our questionnaire is that it counts only the numbers of milk and milk product portions and fails to take into account the actual portion size. Fortunately, the same questions about the amount and frequency of milk product consumption and type of milk products consumed were used throughout the follow-up years, thus ensuring good comparability over time. Moreover, low-lactose milk products were not recorded separately from the milk products with normal lactose content. Despite this limitation, we were able to examine the preference for low-lactose milk and milk products because the subjects were asked to report on low-lactose and milk-free diets. In addition, from the dietary recall data we could calculate the lactose content of the milk products consumed.

The data on the subjects' daily intakes of Ca and lactose were obtained with a detailed 48 h dietary recall. Until the age of 12 years subjects were accompanied by their parents in the dietary recalls and from the age of 15 years they participated in the recalls on their own, which may have influenced the accuracy of the food recording. The estimation of portion sizes may have been better in childhood than in adolescence because children could ask help from their parents and because adolescents eat a larger proportion of the diet outside the home[53]. Ca and lactose intake seemed to decrease in females after the age of 12 years, while in males Ca and lactose intakes increased in adolescence. The most likely explanation for this finding is the underestimation of food intake that is common among adolescent females due to the preoccupation with body weight and image[54]. Furthermore, in both sexes the amount of milk and milk products consumed may reflect the pubertal growth spurt that in average occurs in girls between the ages of 11 and 14 and in boys aged 13–17 years[55].

In conclusion, the C/C$_{-13910}$ genotype was associated with a lower consumption of milk since childhood, predisposing females in particular to inadequate Ca intake. In adult females, the lactose content of milk products consumed was lower, and in both sexes low-lactose and milk-free diets were more common in the C/C$_{-13910}$ genotype than in the other genotypes. In adult females, preference for low-lactose milk and milk products equalised the differences in Ca intake between the genotypes. Thus, in those with the C/C$_{-13910}$ genotype preference for low-lactose milk and milk products may decrease the risk for inadequate Ca intake.

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