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The protective effect of cheese consumption at 18 months on allergic diseases in the first 6 years

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Abstract

Background: The effect of exposure to microorganisms on allergic diseases has been well studied. The protective effect of early food diversity against allergic diseases was previously shown in the PASTURE cohort study. The consumption of cheese, a food potentially rich in microbial diversity, deserves further examination. We aimed to evaluate whether cheese consumption is associated with allergic diseases.

Abbreviations: AD, atopic dermatitis; FA, food allergy; PASTURE, Protection against Allergy—STUdy in Rural Environment.

SN and ADC should be considered as co-first authors.

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Methods: In the PASTURE study (birth cohort in 5 European countries), data on feeding practices, environmental factors, and allergic diseases were collected by questionnaires from birth to 6 years (N = 931). Cheese consumption at 18 months of age was quantified in terms of frequency and diversity (ie, number of consumed types among 6 types: hard pressed, semipressed, soft, blue, fresh cheese, and cheese from the farm). Multiple logistic regressions were performed to evaluate the effect of cheese consumption on atopic dermatitis (AD), food allergy (FA), allergic rhinitis, asthma, and atopic sensitization at 6 years after adjustment for confounders of atopy.

Results: Cheese consumption (vs. nonconsumption) had a significant protective effect on AD (OR = 0.51 [0.29-0.90], P = 0.02) and FA (OR = 0.32, [0.15-0.71], P = 0.004), but no effect on atopic sensitization, allergic rhinitis, and asthma at 6 years. This effect on AD and FA may be related to the diversity of consumed cheeses (OR = 0.64 [0.48-0.85] per cheese type, P = 0.002; OR = 0.55 [0.33-0.92], P = 0.02, respectively).

Conclusion: Although reverse causality cannot totally be ruled out, cheese diversity at 18 months had a protective effect against AD and FA at 6 years in addition to the protective effect of diversity of other foods.

KEYWORDS

allergic disease, atopic dermatitis, cheese, complementary feeding, food allergy

1 | INTRODUCTION

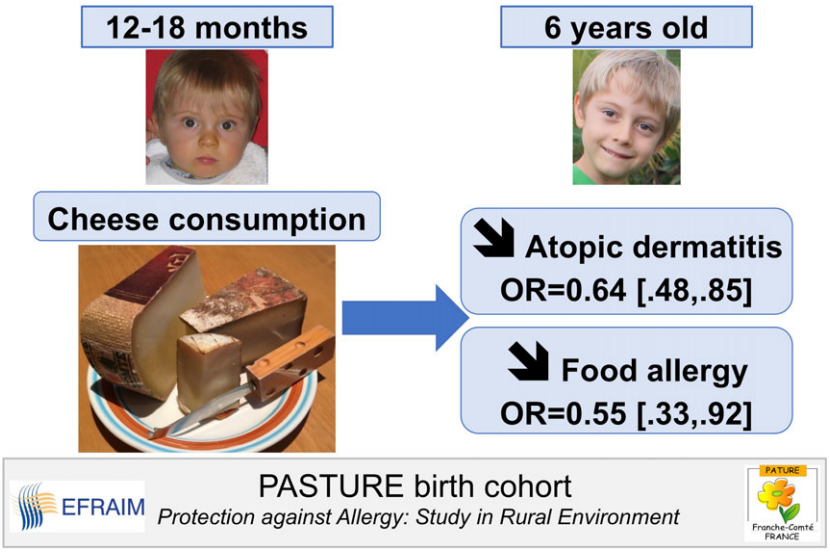
Prenatal and early-life environmental (including dietary) exposures influence immune responses and the development of allergic diseases. Children from rural areas growing up on farms are at a significantly lower risk for developing asthma, allergic rhinoconjunctivitis, and allergic sensitization to inhalant and food allergens than children living in the same rural areas but not growing up in farms. This protective "farm effect" has been associated with contact with livestock, animal feed, and consumption of dairy products.[1-6] An inverse association of consumption of unprocessed cow's milk and the risk of childhood allergic diseases has been observed independently from other farm exposures.[7-9] The independent farm and milk preventive effects may be explained by the quantity and the diversity of microorganisms to which children are exposed to during the first months of life.[10,11]

Immune tolerance to allergens is associated with the quantity and the diversity of microbes that constitute the gut microbiota,[12] and diet is one of the main determinants of the microbial multiplicity of the gastrointestinal tract.[13,14] The birth cohort study "Protection against Allergy—STUdy in Rural Environment (PASTURE)" offered the opportunity to evaluate the effects of early food consumption on the development of allergic diseases.[15] Previous findings on the protective effect of an increased diversity of complementary foods in infant's diet on atopic dermatitis (AD), food allergy, and asthma are in agreement with the hypothesis that exposure to a variety of food antigens and microbes during an early-life time window may be crucial for development of immune tolerance.[16,17] Fermented dairy products and especially traditional cheeses contain a considerable quantity and diversity of microbes, especially bacteria, but also yeasts and molds.[18] We thus longitudinally evaluated whether cheese consumption at the age of 18 months, in terms of frequency and diversity, was associated with the development of AD, asthma, rhinitis, food allergy (FA), and atopic sensitization, using available data from questionnaires up to the age of 6 years. In addition, we assessed the effect of cheese consumption independently from the introduction of other complementary foods.

2 | METHODS

2.1 | Study design and population

The PASTURE study is a prospective birth cohort involving children from rural areas in 5 European countries (Austria, Finland, France, Germany, and Switzerland), designed to evaluate risk and preventive factors for atopic diseases.[15] Pregnant women were recruited during the third trimester of pregnancy between August 2002 and March 2005 and divided into two groups. Women who lived on family-run farms where any kind of livestock was kept were assigned to the farm group. Women from the same rural areas not living on a farm constituted the reference group. In total, 1133 children were included. The study was approved by the local research ethics committees in each country, and written informed consent was obtained from all parents.



GRAPHICAL ABSTRACT

Data from the PASTURE cohort were used to evaluate the effect of cheese consumption at 18 months of age against allergic diseases up to 6 years of age. Multivariate analyses adjusted for major confounders showed a protective effect of cheese consumption and variety against atopic dermatitis and food allergy, but not on allergic rhinitis, asthma, and sensitization to allergens. Reverse causality cannot be totally ruled out but is unlikely. Hypothesis: The variety of cheese consumed in early life could influence the immune system, through microbial components and by anti-inflammatory compounds (short-chain fatty acids)

2.2 | Definitions

Questionnaires were administrated during interviews or self-administered to the mothers within the third trimester of pregnancy and when the children were 2, 12, 18, 24 months of age and then yearly up to the age of 6 years. Children were defined as having asthma when the parents reported that the child had either a doctor diagnosis of asthma at least once or at least 2 episodes of doctor-diagnosed obstructive bronchitis in the last 12 months between 3 and 6 years of age, independently from diagnosis reported in the first 3 years. Food allergy was defined when the parents reported up to 6 years of age that the child had had at least once a doctor diagnosis of food allergy. Allergic rhinitis was defined by the presence of symptoms (itchy, runny or blocked nose, and red itchy eyes) or doctor diagnosis of allergic rhinitis ever up to 6 years of age, built using data reported in the 6-year questionnaire and before. Children were defined having AD when the parents reported that the child had AD diagnosed by a doctor at least once in the questionnaire between 12 months and 6 years, and/or with positive SCORAD score (>0) assessed at the age of 1 year during medical examination. Children with no AD, but missing information at least at one time point, were defined as missing.

Specific IgE antibodies to the following allergens (*D. pteronyssinus*, *D. farinae*, alder, birch, hazel, grass pollen, rye, mugwort, plantain, cat, horse, dog, *Alternaria*, hen's egg, cow's milk, peanut, hazelnut, carrot, and wheat flour) were measured in blood at the age of 1, 4.5, and 6 years. Sensitization was defined as specific IgE level of 3.5 kU/L or more, and as being strongly associated with allergic disease. Sensitization was defined for food allergens and for inhalant allergens separately.

Parents indicated which food items had been given to the child during the last 4 weeks in monthly diaries between the 3rd and 12th months, and in the last 6 months in the questionnaire collected at the age of 18 months, including cheese. To describe the introduction to complementary foods, we used the previously defined food diversity score.[16,17] This score reports the introduction of the major food groups, that is, those introduced in the first year to at least 80% of the children (vegetables or fruits, cereals, bread, meat, cake, and yogurt). Any type of cheese consumption at 18 months was defined as a binomial variable (yes/no). The frequency of consumption of cheese was taken into account (never, less than once a week, one to 6 times per week, and at least once a day). Moreover, since the different brands of cheese consumed by the child at the age of 18 months were reported by the parents, cheese was categorized into 6 types according to cheese families described by Montel et al.[18]: hard pressed, semipressed, soft, blue, fresh cheeses, and cheese from the farm. Cheese diversity score was defined by the number of cheese types consumed at 18 months and considered as a continuous variable in the analysis.

Farmer children were defined as children who were living on a farm where livestock was held and whose family ran the farm, according to parental reports. Information on the parental atopic status, maternal education, smoking during pregnancy, mode of delivery, birthweight, gestational age, sex, number of siblings, and the duration of breastfeeding (either exclusively or not) was recorded in questionnaires during pregnancy, 2 months after birth, and 1 year of age. Parental history of allergies was defined as at least one parent ever having asthma, allergic rhinitis, or AD (none, at least one).

2.3 | Statistical analysis

Data analysis was conducted using SAS software for Windows version 9.4 (SAS Institute, Inc., Cary, NC). Differences in cheese consumption (yes/no) and consumption frequency among children in terms of farmer status, center, sex, siblings, parental atopy, breastfeeding, and maternal education were analyzed with chi-square tests, and differences in cheese diversity score were analyzed with ANOVAs.

Logistic regressions analysis was used to determine whether cheese consumption (yes/no), cheese consumption frequency, and cheese diversity were associated with a protective effect against AD, FA, allergic rhinitis, asthma, food sensitization, and inhalant sensitization. Some confounders were forced in all models: center, farmer status, and parental antecedent of atopy (0, 1, or 2). Other potential confounders were tested for each outcome in bivariate analyses: maternal smoking during pregnancy (no, yes), mode of delivery, mother living in the farm, mother working at the farm after birth, maternal education, presence of siblings, sex, gestational age, birthweight, breastfeeding duration, and score of food diversity at 12 months.[16,17] Only variables with likely associations with each outcome ($P < 0.20$) were included in multivariate models. Thus, some variables were not considered further (presence of siblings, maternal smoking during pregnancy, gestational age, and birthweight). To take collinearity into account, the absence of associations among the quantitative variables from this list (gestational age, birthweight, score of food diversity at 12 months, and cheese diversity score at 18 months) was confirmed by analyzing the variance inflation factor (VIF<5). Finally, multivariate linear logistic regressions were performed to evaluate the association between cheese consumption/diversity at 18 months and AD, FA, allergic rhinitis, asthma, and food or inhalant IgE (with the 3.5 kU/L cutoff), with adjustment for the potential confounding factors identified as described above, and for center, farmer status, and parental atopy. Since the analysis for each outcome answers a different question, no adjustment of *P*-values for multiple testing was performed,[19] and significance was set at $P < 0.05$.

Additional analyses were conducted to evaluate whether the effect of cheese consumption/diversity was still significant after adjustment for farm/raw milk consumption at 12 months. In order to take potential reverse causality effects into account, further analyses were conducted after exclusion of infants who presented diagnosis of FA at 12 months or of AD at 18 months, whose parents may have excluded some foods from their diet, or after exclusion of children who were not given any milk, milk product, or yogurt at 12 months.

Results are presented as OR [95% confidence interval].

3 | RESULTS

3.1 | Sample selection

At baseline, 1133 pregnant women were recruited. Among the infants, some had missing data regarding cheese consumption at 18 months or allergic diseases at 6 years, leaving data for a maximum of 931 infants.

3.2 | Sample characteristics, cheese consumption, and prevalence of allergic diseases

Among the 931 included children, 47.8% were farmer's children (Table 1). Only 7.5% of the children did not consume any cheese at 18 months. As shown in Table S1, most children (53.9%) consumed cheese 1-6 times per week at 18 months. On average, children received 1.2 ± 0.6 cheese types at 18 months (Table 1); most children (66.2%) had received one cheese type, 23.2% 2 cheese types, and 2.6% 3 or more cheese types. The most consumed (here by at least 10 children) cheese types in each country are reported in Table 2. Gouda was the most consumed cheese in Austria (n = 60 consumers), Tilsiter in Switzerland (n = 64), Comté in France (n = 140), Gouda in Germany (n = 37), and Edam in Finland (n = 116).

Differences between centers were observed regarding the cheese diversity score (Table 1): It was higher in France than in the other countries. The cheese diversity score did not differ between children of farmers and nonfarmers ($P = 0.26$); it was not associated with sex, number of siblings, and parental atopy. It was lower when breastfeeding duration increased, and higher in children of mothers with mid-high education attainment. Daily cheese consumption was more frequent in French and Finnish children, and in children of mothers with mid-high and high education (Table S1).

The cheese diversity score at 18 months was not correlated to the food diversity score at 12 months ($r = 0.02$, $P = 0.48$). The food diversity score at 12 months was marginally higher in consumers than nonconsumers of cheese (5.5 ± 0.9 vs. 5.1 ± 1.4, $P = 0.05$, Table 1) and did not vary according to cheese consumption frequency at 18 months (Supplemental Table 1).

3.3 | Association between cheese consumption at 18 months and allergic diseases and atopic sensitization at 6 years

Any consumption of cheese at 18 months was associated with significantly reduced risk of AD at 6 years (OR, 0.51 [0.29-0.90], $P = 0.02$), of FA (OR, 0.32 [0.15-0.71], $P = 0.004$) and a reduced risk, albeit nonsignificant, of allergic rhinitis (OR, 0.51 [0.22-1.19], $P = 0.12$), asthma (OR, 0.61 [0.27-1.40], $P = 0.24$), and sensitization to food allergens (OR, 0.50 [0.21-1.19], $P = 0.12$) and inhalant allergens (OR, 0.59 [0.29-1.23], $P = 0.16$) (Table 3, model 1). Notably, these models include adjustment for potential confounders including food diversity score at 12 months. Even after adjustment on raw milk consumption, the reduced risk of AD was significant (OR, 0.51 [0.29-0.90], $P = 0.02$, Table 3, model 2) as well as the reduced risk of FA (OR, 0.32 [0.14-0.69], $P = 0.004$).

3.4 | Association between cheese diversity and allergic diseases and atopic sensitization

Because most children ate cheese, we also considered the effect of cheese diversity at 18 months. This analysis revealed a decreased OR of AD (0.64, [0.48-0.85]; $P = 0.002$; Table 4, model 1) and of FA (0.55, [0.33-0.92]; $P = 0.02$) with increasing cheese diversity, but no

TABLE 1 Characteristics of the study population and cheese diversity at 18 mo

	Cheese consumption							Cheese diversity score	
	All		No		Yes				
Characteristics	n	%	n	%	n	%	*P* value[a]	Mean ± SD	*P* value[b]
All	931	100	70	7.5	861	92.5		1.21 ± 0.62	
Farmer									
Yes	445	47.8	32	7.2	413	92.8	0.8	1.23 ± 0.62	0.26
No	486	52.2	38	7.8	448	92.2		1.18 ± 0.62	
Center									
Austria	179	19.2	17	9.5	162	90.5	**0.0051**	1.19 ± 0.68	**<0.0001**
Switzerland	203	21.8	25	12.3	178	87.7		1.04 ± 0.61	
France	167	17.9	8	4.8	159	95.2		1.61 ± 0.63	
Germany	218	23.4	15	6.9	203	93.1		1.21 ± 0.61	
Finland	164	17.6	5	3.1	159	96.9		1.02 ± 0.28	
Sex									
Girls	458	49.23	32	7	426	93	0.62	1.24 ± 0.64	0.14
Boys	471	50.7	37	7.9	434	92.1		1.18 ± 0.60	
Siblings									
Yes	589	63.3	44	7.5	545	92.5	1	1.21 ± 0.60	0.93
No	342	36.7	26	7.6	316	92.4		1.20 ± 0.62	
Parental atopy									
None	429	46.3	27	6.3	402	93.7	0.18	1.23 ± 0.61	0.32
One parent	400	43.2	37	9.3	363	90.7		1.20 ± 0.65	
Both parents	97	10.5	5	5.2	92	94.8		1.12 ± 0.52	
Breastfeeding									
Never	90	9.8	5	5.6	85	94.4	0.82	1.41 ± 0.73	**0.0015**
<3 mo	147	15.9	13	8.8	134	91.2		1.23 ± 0.64	
3-<7 mo	252	27.3	21	8.3	231	91.7		1.23 ± 0.62	
7-<10 mo	198	21.5	13	6.6	185	93.4		1.19 ± 0.60	
≥10 mo	235	25.5	16	6.8	219	93.2		1.09 ± 0.54	
Maternal education									
Low	154	16.5	23	14.9	131	85.1	**0.0008**	1.12 ± 0.67	**0.0096**
Mid	398	42.8	31	7.8	367	92.2		1.18 ± 0.62	
Mid-high	274	29.4	11	4	263	96		1.31 ± 0.61	
High	105	11.3	5	4.8	100	95.2		1.18 ± 0.53	
Food diversity score	917[c]	5.4 ± 1[d]	66	5.1 ± 1.4	851	5.5 ± 1	0.05[b]		
Eviction of milk, yogurt, and dairy products other than cheese at 12 mo									
Yes	96	10.8	14	14.6	82	85.4	**0.0048**	1.00 ± 0.58	**0.0005**
No	791	89.2	48	6.1	743	93.9		1.24 ± 0.61	

[a]Based on chi-square test.
[b]Based on ANOVAs.
[c]n = 14 missing values for this variables.
[d]mean ± SD.
Boldface values stand for significant effect ($P < 0.05$).

statistically significant association with allergic rhinitis ($P = 0.78$), asthma ($P = 0.84$), inhalant allergen sensitization ($P = 0.33$), and food allergen sensitization ($P = 0.29$). These effects were similarly observed after adjustment for raw milk consumption at 12 months (Table 4, model 2).

The analysis of the effect of cheese consumption frequency (Table S2, models 1 and 2) confirmed that children who never ate cheese were more at risk of FA than children who consumed cheese 1 to 6 times per week ($P = 0.03$), and showed that children who ate cheese never or less than once a week were more at risk of rhinitis

TABLE 2 Names and types of cheese most frequently consumed by participants in each country

Cheese types	Austria[a]	Switzerland	France	Germany	Finland
Hard pressed	Gouda (60) Emmentaler (40)	Appenzeller (51) Emmentaler (25) Alpkäse (24) Greyerzer (22) Hartkäse (17) Halbhartkäse (17)	Comté (140) Morbier (30) Emmental (17)	Gouda (37) Emmentaler (36) Hartkäse (25) Bergkäse (16) Edamer (12)	Edam (116)
Semipressed	Tilsiter (25)	Tilsiter (64)		Tilsiter (12)	Oltermanni (18) Kermajuusto (10)
Soft	Butterkäse (18)	Weichkäse (11)	Vache qui rit (62) Camembert (60) Cancoillotte (40) Brie (11) Mont d'or (10)	Butterkäse (21) Camembert (19)	
Fresh cheeses	Streichkäse (11)		Kiri/Streichkäse (51)	Frischkäse (27) Streichkäse (11)	

[a]In brackets: number of consuming children at 18 mo of age, for the cheeses consumed by at least 10 children. No specific cheese is reported for the blue and farm types because they were consumed by less than 10 children in all countries.

than children who consumed cheese 1 to 6 times per week ($P = 0.03$ and $P = 0.007$, respectively).

3.5 | Evaluation of the effect of potential reverse causality

In order to take into account reverse causality, similar analyses were conducted after exclusion of the children who were doctor-diagnosed any FA within the first 12 months (Tables 3, 4, models 3) or AD within the first 18 months (Tables 3, 4, models 4). In these analyses, the protective effect of cheese diversity at 18 months against AD was still observed after exclusion of children diagnosed with food allergies at 12 months (OR = 0.70 [0.51-0.91]; $P = 0.01$; Table 4, model 3) but not after exclusion of the 165 children who already had an AD at 18 months (OR = 0.68 [0.44-1.04]; $P = 0.08$; Table 4 model 4). Regarding FA at 6 years, the odds ratios were still in favor of a protective effect but were not significant anymore.

Could the observed effect be due to the children who were not fed any dairy products other than cheese, milk, yogurt or whose parents consciously avoided dairy products at 12 months? As described in Table 1 and Supplemental Table 1, such a consumption pattern is only partly overlapping with the absence of cheese consumption. After exclusion of these children (n = 48), the protective effect of cheese diversity on AD was still observed (OR = 0.71, [0.52-0.98], $P = 0.04$; Table 4, model 5), but the protective effect of cheese consumption or diversity on FA was not significant anymore (Tables 3, 4, models 5).

4 | DISCUSSION

Our data suggest that an increased diversity of cheese types consumed at 18 months might be associated with a protective effect on the development of AD and FA at the age of 6 years. Remarkably, this effect was observed even after adjustment for potential confounders which were previously shown to be associated with a protective effect against the development of allergic diseases, such as parental atopy, maternal contact with farm animal during pregnancy,[20] farm or raw milk consumption, and food diversity during the first year.[16,17]

To the best of our knowledge, this is the first report on the effect of early cheese consumption on the development of allergic diseases.

The strengths of this study are the prospective design and the detailed data collection of feeding practices in first years of life, including consumption of different cheeses, which made it possible to analyze the effect of the frequency and diversity of cheese consumption at 18 months of age on the development of allergic diseases. In the same cohort, an increased diversity of food introduced in children's diet within the first year of life was associated with a protective effect on the development of AD, asthma, and FA up to 6 years, and on sensitization to food allergens at 4.5 or 6 years.[16,17] In our study, it is noticeable that the protective effect of an increased cheese diversity at 18 months is additional to the protective effect of the diversity of other food groups consumed in the first year (vegetables or fruits, cereals, bread, meat, cake, yogurt, and raw milk).[16,17] This protective effect was found only for AD and FA but not for asthma, hay fever or atopic sensitization. AD and FA are the first symptoms of allergy in young children, while asthma and hay fever appear later. However, there is a tendency for a protective effect of cheese consumption on asthma and hay fever (Table 4), and the absence of significant effect may be explained by the multifactorial origin of asthma and rhinitis.

One major concern with the association between feeding practices and atopic diseases is the potential bias due to reverse causality. Diagnosis of AD and/or FA may have encouraged parents to exclude cheese from their child's diet. However, most of the children (77.4%)

TABLE 3 Associations between cheese consumption (yes/no) at 18 mo and allergic diseases and atopic sensitization at 6 yr

	Model 1			Model 2			Model 3			Model 4			Model 5		
	n	OR	95% CI	n	OR	95% CI	n	OR	95% CI	n	OR	95% CI	n	OR	95% CI
Atopic dermatitis[a]	258/839	**0.51**	**0.29-0.90**	**258/839**	**0.51**	**0.29-0.90**	241/820	0.63	0.34-1.17	93/674	0.61	0.26-1.45	211/722	0.71	0.35-1.44
Doctor-diagnosed food allergy[b]	68/843	**0.32**	**0.15-0.71**	**68/843**	**0.32**	**0.14-0.69**	48/823	0.65	0.21-1.96	33/697	0.38	0.12-1.24	48/724	1.13	0.25-5.05
Allergic rhinitis[c]	64/870	0.51	0.22-1.19	64/870	0.50	0.22-1.18	59/856	0.95	0.32-2.84	47/725	0.76	0.25-2.34	52/751	1.004	0.29-3.54
Asthma[d]	73/837	0.61	0.27-1.40	73/837	0.61	0.27-1.4	65/822	1.11	0.38-3.27	54/701	0.51	0.19-1.32	54/722	0.78	0.26-2.36
Sensitization to food allergens (cut off 3.5 KU/L) at 6 yrs[e]	5/707	0.50	0.21-1.19	48/698	0.65	0.25-1.71	46/692	0.83	0.29-2.36	38/582	0.84	0.27-2.65	40/613	1.08	0.30-3.91
Sensitization to inhalant allergens (cut off 3.5 KU/L) at 6 yrs[f]	109/704	0.59	0.29-1.23	105/695	0.74	0.34-1.6	104/689	0.87	0.37-2.03	80/579	0.75	0.3-1.88	93/610	0.93	0.35-2.45

For all diseases, the modeled probability is the probability to observe the disease.
Boldface values are significant ($P < 0.05$).
Model 1: adjustment for centre, farmer, and parental atopy plus other variables as described below.
Model 2: model 1 + additional adjustment for raw milk consumption at 12 mo.
Model 3: model 1 + exclusion of infants with doctor-diagnosed FA at 1 yr.
Model 4: model 1 + exclusion of infants with AD at 18 mo.
Model 5: model 1 + exclusion of infants who were non consumers of dairy products other than cheese, yogurt, and milk at 12 mo.
[a]Also adjusted for maternal education, mode of delivery, mother working on the farm after birth, breastfeeding duration, and score of food diversity at 12 mo.
[b]Also adjusted for mother working on the farm after birth, birthweight, and score of food diversity at 12 mo.
[c]Also adjusted for sex, mother living on the farm, other children in family, mother working on the farm after birth, and score of food diversity at 12 mo.
[d]Also adjusted for sex, mother living on the farm, mother working on the farm after birth, and score of food diversity at 12 mo.
[e]Also adjusted for sex, mother working on the farm after birth, and maternal education.
[f]Also adjusted for sex, other children in family, maternal education, birthweight, and breastfeeding duration.

TABLE 4 Associations between cheese diversity score at 18 mo and allergic diseases and atopic sensitization at 6 yr

	Model 1			Model 2			Model 3			Model 4			Model 5		
	n	OR	95% CI	n	OR	95% CI	n	OR	95% CI	n	OR	95% CI	n	OR	95% CI
Atopic dermatitis[a]	245/788	**0.64**	**0.48-0.85**	245/788	**0.64**	**0.48-0.85**	229/770	**0.7**	**0.51-0.91**	92/635	0.68	0.44-1.04	**203/678**	**0.71**	**0.52-0.98**
Doctor-diagnosed food allergy[b]	62/790	**0.55**	**0.33-0.92**	62/790	**0.54**	**0.32-0.91**	43/771	0.68	0.37-1.24	31/655	0.86	0.42-1.74	44/678	0.82	0.45-1.51
Allergic rhinitis[c]	60/818	0.93	0.56-1.54	60/818	0.93	0.56-1.53	55/804	1.17	0.70-1.94	44/682	1.26	0.72-2.21	48/705	1.13	0.66-1.94
Asthma[d]	67/786	0.96	0.61-1.5	67/786	0.96	0.61-1.51	59/771	1.13	0.72-1.79	50/659	0.82	0.48-1.40	50/677	1.12	0.68-1.85
Sensitization to food allergens (cut off 3.5KU/l) at 6 yrs[e]	48/667	0.75	0.44-1.29	46/658	0.83	0.48-1.44	44/652	0.91	0.53-1.57	37/549	0.85	0.46-1.57	38/578	1.09	0.61-1.96
Sensitization to inhalant allergens (cut off 3.5KU/l) at 6 yrs[f]	103/664	0.9	0.61-1.31	99/655	0.95	0.65-1.40	98/649	1.03	0.70-1.51	76/546	0.97	0.62-1.51	87/575	1.07	0.70-1.63

Boldface values are significant ($P < 0.05$).

Model 1: adjustment for centre, farmer, and parental atopy plus other variables as described below.

Model 2: model 1 + adjustment for raw milk consumption at 12 mo.

Model 3: model 1 + exclusion of infants with doctor-diagnosed FA at 1 yr.

Model 4: model 1 + exclusion of infants with AD at 18 mo.

Model 5: model 1 + exclusion of infants who were non consumers of dairy products other than cheese, yogurt, and milk at 12 mo.

[a]Also adjusted for maternal education, mode of delivery, mother working on the farm after birth, breastfeeding duration, and score of food diversity at 12 mo.

[b]Also adjusted for mother working on the farm after birth, birthweight, and score of food diversity at 12 mo.

[c]Also adjusted for sex, mother living on the farm, other children in family, mother working on the farm after birth, and score of food diversity at 12 mo.

[d]Also adjusted for sex, mother living on the farm, mother working on the farm after birth, and score of food diversity at 12 mo.

[e]Also adjusted for sex, mother working on the farm after birth, and maternal education.

[f]Also adjusted for sex, other children in family, maternal education, birthweight, and breastfeeding duration.

who had never eaten cheese at 18 months had not avoided milk, milk products, or yogurt at 12 months. In the analyses performed in a subgroup of children excluding those with FA or AD in the first year, the observed associations were not significant anymore, which does not make it possible to exclude reverse causality. The low number of remaining cases after exclusion for early onset leads to a loss of power. Most of children with AD at 6 years (63.9%) already had AD at 18 months. Furthermore, an early occurring phenotype of AD is known to be associated with FA and food sensitization and an early occurring then persistent phenotype of AD is associated with a higher risk of developing asthma later in life.[21] The exclusion of early phenotypes of AD thus leads to the loss of children most at risk of allergies and makes the associations not significant anymore.

Regarding the hypotheses that could explain a protective effect on the development of allergic diseases, the first one is the microbial diversity of cheeses.[18,22] Cheeses are one of the oldest fermented foods created by humans. Worldwide, there are about 1000 distinct types of cheese. Each manufacturing unit has a specific house microflora, dependent on the environmental conditions. The classification of cheese presently used and previously described by Montel et al. [18] is based on the mode of preparation, the type of rennet, starters, and ripening. However, raw milk or pasteurized milk may be used in each type, and the exact microbial composition of each type is unknown. The composition of microbial communities varies according to cheese variety with a great influence of the cheese manufacturing process.[22] Moreover, the importance of rind type and moisture on the structure of cheese microbial communities was highlighted.[23] Cheeses are inoculated with standard and marketed starter cultures and there is a secondary microbiota composed of bacteria, yeasts, and molds, originated from milk and the environment that is usually recovered from the cheese's surface and core.[24] Some of these bacterial strains have been selected as probiotics.

During the past 10 years, several studies have shown that a rich diversity of gut microbiota in early childhood has a protective effect on the development of allergic diseases. According to Abrahamsson et al, low intestinal microbial diversity during the first month of life is associated with subsequent AD and asthma.[25] Bisgaard et al showed that reduced bacterial diversity of the infant's intestinal flora was associated with increased risk of allergic sensitization, allergic rhinitis, and peripheral blood eosinophilia, but not asthma or AD, in the first 6 years.[26]

In relation to its rich microbial diversity, cheese could induce a rich diversity of gut microbiota. In rats, the consumption of cheese whey protein increases the *Lactobacilli* and *Bifidobacteria* counts, and the fecal mucin secretion.[27] In human microbiota-associated rats, camembert consumption led to a potentially beneficial influence on the intestinal metabolism.[28] Another hypothesis to explain the protective effect of cheese consumption on allergic diseases is a potential anti-inflammatory effect. Several studies focused on the relationship between consumption of fermented dairy products and gut inflammation. Some specific probiotic strains have an anti-inflammatory effect in mice and humans.[29] The consumption of a *Lactobacillus helveticus*-containing cheese significantly inhibited immune cell proliferation and proinflammatory cytokine production,[30] suggesting that this strain has beneficial immunoregulatory properties. Short-term feeding of mice with Roquefort cheese led to the inhibition of the migration of peritoneal leukocytes, to the reduction in neutrophil count, and a relative increase in peritoneal macrophages, suggesting that ingestion of Roquefort could promote regenerative processes at the site of inflammation.[31]

This potential anti-inflammatory effect has also been related to levels of metabolites produced by intestinal microbiota after dairy product consumption.[32,33] More specifically, short-chain fatty acids such as propionate and butyrate, found in cheese,[34] might present such anti-inflammatory properties.[35] In a recent study, differences in serum fatty acid pattern seem to be related to subsequent allergy development,[36] and ω-3 fatty acids were found to contribute to the protection against asthma in the PASTURE cohort.[37]

All these interactions between dietary factors and gut microbiota reinforce the hygiene hypothesis,[38] which could be renamed "microbiota hypothesis," as recently suggested.[39] Further data must be collected to strengthen the understanding of the cheese consumption effect on a putative modification of gut microbiota. For instance, timing of exposure associated with higher protection must be qualified. The timing of cheese introduction may be a bit late to alter asthma development given the current knowledge on the effect of environment-driven microbiota alteration on asthma in early months.[40] This may explain that cheese consumption did not affect similarly all atopic outcomes. Furthermore, several studies on the effect of gut microbiota diversity on allergic diseases found an association with one or two atopic outcomes but not on all the atopic outcomes.[26] However, the composition of the protective gut microbiota remains unclear and could be different according to the type of allergic disease.[41-43]

5 | CONCLUSION

Cheese diversity at 18 months has a protective effect against AD and FA at 6 years in addition to and independently from the protective effect of a diversity of other foods. These results are in line with the "hygiene" hypothesis with a probable increase in the diversity of gut microbiota. However, the reverse causality cannot be totally excluded. Randomized control trials are needed to confirm our results, to determine whether diversity or frequency of cheese consumption is the most important protective factor, and which is the most effective window of exposure to cheese to help protect against early AD onset. In parallel, analyses of gut microbiota of cheese eaters could help to understand the underlying mechanisms.

DECLARATION OF CONFLICT OF INTEREST

The authors report no conflict of interest.

AUTHOR CONTRIBUTIONS

JCD, SN, DAV, and ADC conceived and designed the study. MLC, CR, VK, MLD, AMK, PK, JP, RL, ESH, HR, CBF, JR, DAV, EVM, and JCD collected the data. EK, SN, and ADC performed the statistical analyses and interpreted the results. SN, ADC, and EK drafted the manuscript. SN, ADC, CR, VK, EK, AMK, PK, JP, RL, HR, CBF, JR, DAV, EVM, and JCD approved the final version of the manuscript.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

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