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ORIGINAL ARTICLE

Maternal intake of fatty acids and their food sources during lactation and the risk of preclinical and clinical type 1 diabetes in the offspring

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Abstract
Aims We examined maternal dietary intake of fatty acids and foods which are sources of fatty acids during lactation and whether they are associated with the risk of preclinical and clinical type 1 diabetes in the offspring.
Methods The subjects comprised a cohort of 2,939 mother–child pairs from the prospective Type 1 Diabetes Prediction and Prevention Study. Composition of maternal diet during the third month of lactation was assessed by a validated food frequency questionnaire. Among the children with HLA-conferred susceptibility to type 1 diabetes, 172 developed preclinical and 81 clinical diabetes. Average follow-up for preclinical type 1 diabetes was 7.5 years (range 0.2–14.0 years) and for clinical type 1 diabetes 7.7 years (0.2–14.0 years).
Results Maternal intake of fatty acids during lactation was not associated with the risk of type 1 diabetes in the offspring. After adjusting for putative confounders, maternal total consumption of red meat and meat products during lactation was associated both with increased risk for preclinical [hazard ratio (HR) 1.19, 95 % CI 1.02–1.40, $p = 0.038$] and clinical type 1 diabetes (HR 1.27, 95 % CI

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1.06–1.52, $p = 0.025$). In particular, consumption of processed meat products showed an association with increased risk for type 1 diabetes (HR 1.23, 95 % CI 1.02–1.48, $p = 0.045$). Maternal use of vegetable oils was associated with increased risk for preclinical type 1 diabetes (HR 1.21, 95 % CI 1.03–1.41, $p = 0.023$).

Conclusions Maternal consumption of red meat, especially processed meat, during lactation may increase the risk of type 1 diabetes.

Keywords Lactation · Fatty acid · Type 1 diabetes · Child · Cohort

Introduction

Prenatal and early infant nutrition modify development and maturation of gut and immune responses in the fetus and in the infant [1]. Maternal nutrition during pregnancy and lactation may play a role in the development of type 1 diabetes, which is thought to be influenced by gut-associated autoimmune inflammatory reactions [2, 3]. Fatty acids have been shown to affect immune responses and inflammatory reactions [4, 5]. A few epidemiological studies have indicated that the intake of n-3 fatty acids or the use of cod liver oil during early childhood could be protective against autoimmunity or clinical type 1 diabetes [6–8]. During early infancy, breast milk is an important source of n-3 fatty acids. Maternal fatty acid stores and maternal diet during lactation affect strongly the fatty acid composition of breast milk and thus the fatty acid availability and status in infancy [9–13].

As far as we know, there are no studies available on the associations between maternal food consumption or dietary fatty acid composition during lactation and the risk of type 1 diabetes. The accumulated evidence on the association of breastfeeding with type 1 diabetes is controversial [14]. Case–control findings have indicated that short duration of breastfeeding and early age at the introduction of cow's milk-based formulas is associated with an increased risk for preclinical or clinical type 1 diabetes [15]. However, most of the prospective cohort studies have not shown any association between breastfeeding and risk of autoimmunity or type 1 diabetes [16–19], while some have found a protective effect [20, 21].

We previously observed that maternal consumption of fresh milk and cheese and low-fat margarines and intake of fresh milk proteins during pregnancy were associated with lower risk of preclinical or clinical type 1 diabetes, while the consumption of sour milk products and intake of sour milk proteins were associated with increased risk of preclinical type 1 diabetes [22]. The aim of the present study was to assess the intakes of different fatty acids and their food sources among lactating Finnish women and the associations with the development of preclinical and clinical type 1 diabetes in their offspring.

Participants and methods

The participants comprised a cohort of 2,939 mother–child pairs from the prospective Finnish Type 1 Diabetes Prediction and Prevention (DIPP) study. Data included 26 twin pairs, and thus, maternal dietary data are provided from 2,913 mothers who breastfed their babies during the first 3 months. The children were born between 1998 and 2004. In the DIPP study, infants with genetic susceptibility to type 1 diabetes were monitored for the appearance of signs of beta-cell autoimmunity and clinical type 1 diabetes. The average follow-up time for preclinical type 1 diabetes was 7.5 years (range 0.2–14.0 years), and for clinical type 1 diabetes, follow-up time was 7.7 years (0.2–14.0 years). Details of the genetic screening methods [23, 24], enrollment criteria [24] and the analysis of autoantibodies [25] have been described in detail previously. Socio-demographic factors, e.g., maternal education and familial diabetes, were registered by a structured questionnaire completed by the parents after delivery.

Definition of outcome

We defined preclinical type 1 diabetes as being repeatedly positive for islet cell autoantibodies in combination with repeated positivity for one or more of the other autoantibodies: insulin autoantibodies, autoantibodies to the 65 kDa isoform of glutamic acid decarboxylase and autoantibodies to the tyrosine phosphatase-related islet antigen 2. Among the 81 children progressing to clinical type 1 diabetes, 67 had been repeatedly positive for islet cell autoantibodies and at least one other autoantibody. However, seven of the remaining 14 children who progressed to type 1 diabetes had or had had one or more autoantibodies before or at the time of diagnosis. Only seven children who developed type 1 diabetes had been persistently seronegative before diagnosis. Thus, preclinical type 1 diabetes endpoint is defined as the first occurrence of either: (1) repeated positivity for islet cell autoantibodies in combination with one or more of the autoantibodies analyzed or (2) clinical type 1 diabetes. By September 2012, 172 children have developed preclinical type 1 diabetes, and by January 2013, 81 have progressed to clinical diabetes. The mean age for developing preclinical endpoint was 4.4 years (range 0.5–13.0 years), and the mean age for disease diagnosis was 6.1 years (range 1.3–13.2 years).

Assessment of maternal nutrition

Mothers completed a validated 181-item semiquantitative food frequency questionnaire (FFQ) during the third month of lactation. The FFQ contained questions enquiring about the frequency (number of times per day, week or month or not at all) and the amount of foods consumed, in units of common serving sizes. For instance, the consumption of fish was evaluated by 13 questions, so that lean and fatty fish were asked separately. Consumption of meat was asked by 25 questions which four questions concerned cold cuts and one question sausages. Furthermore, the consumption of fat used on bread was evaluated by six separate questions including the number of bread slices as well as the type and amount of fat spread on the bread. In addition, the FFQ contained questions related to the type of fat used in cooking, baking and in salad dressings as well as questions related to the extent of home baking. The individual type and quantity of fat were taken into account when calculating the food and nutrient intake. Each food row in the FFQ was transformed into ingredients according to an individual recipe based on the average food consumption of Finnish women of fertile age. The daily nutrient intakes were calculated using the updated Finnish Food Composition Database and in-house software of the National Institute for Health and Welfare. The content of the FFQ and data processing has been described in detail [26].

Child's serum fatty acid composition

In order to analyze correlations between maternal fatty acid intake and child's serum fatty acid composition, we analyzed non-fasting serum total fatty acid composition from a subset of 135 seronegative children who were exclusively breastfed until the serum sample nearest to the age of 3 months was obtained [mean age at the time of serum sample was taken 3.0 months (range 2.0–4.3 months)]. The definition of exclusively breastfeeding was that breast milk has been the only drink and food which infant is exposed to. The only exceptions are water and vitamin or mineral supplementation. Serum total fatty acid composition was analyzed by gas chromatography. Handling of serum samples and gas chromatography method was described more detail previously [27]. The fatty acid composition was expressed as a percentage of total fatty acids in serum.

Statistical analyses

The endpoint of preclinical type 1 diabetes is interval censored and possibly dependent among siblings. Therefore, a piecewise linear log-hazard survival model was used to analyze the associations of fatty acid intake and food consumption with the risk of preclinical type 1 diabetes. Linear log-hazards in the intervals 0–1.99, 2–3.99 and ≥4 years were assumed. Observation intervals beyond positivity did not contribute to the analysis. The models were fitted using maximum likelihood in SAS PROC NLMIXED, with standard errors of the estimates derived from the observed information matrix. The endpoint of clinical type 1 diabetes is not interval censored; therefore, the associations of fatty acid intake and food consumption with the risk of clinical disease were analyzed using Cox proportional hazard regression.

Fatty acid variables were adjusted for energy intake by the residual method [28] after logarithmic transformation. Food variables were adjusted for energy intake by adding energy intake as a covariate to the survival models. Depending on the distribution, fatty acid and food variables were used as continuous or categorical variables in the analyses. Some food variables were dichotomized because of high proportion of non-users (for high-fat margarines 59 %, for low-fat margarines 71 %). The possible confounding by background characteristics (vocational education of the mother, genetic risk group, familial diabetes and exclusive duration of breastfeeding) was taken into account by adding background variables as covariates to the survival model. SAS version 9.2 (SAS Institute, Cary, NC, USA) was used in the analyses. Statistical significance was taken as <5 %.

To assess correlations between maternal fatty acid intake during the third month of lactation and child's serum fatty acid status at 3 months of age for exclusively breastfed children, we used Spearman's correlation coefficients. Correlations between maternal fatty acid and food intake during pregnancy and lactation were also assessed using Spearman's correlation coefficients.

Results

Of the children who were included in the analysis, 97 % were breastfed at the age of 3 months. Proportion of exclusively breastfed infants was 43.2 % and proportion of partially breastfed babies was 56.8 %. The mean (SD) duration of exclusive breastfeeding was 1.8 (1.7) months, while the mean duration of overall breastfeeding was 7.4 (5.1) months. Maternal mean intake of total fatty acids was 32.8 % of energy intake (*E*%), saturated fatty acids (SFA) 14.9 *E*%, monounsaturated fatty acids (MUFA) 12.5 *E*%, polyunsaturated fatty acids (PUFA) 4.9 *E*%, n-6 PUFA 3.7 *E*% and n-3 PUFA 1.1 *E*%. Maternal median daily intake of fatty acids from foods and median consumption of food sources during lactation are presented in Tables 1 and 2.

Maternal intake of pentadecanoid acid, stearic acid, conjugated linoleic acid, alphalinolenic acid (ALA),

Table 1 Maternal daily intake of fatty acids during lactation in the years 1998–2004 and correlation coefficients between maternal intake of fatty acids during lactation and the child's serum fatty acid concentration at 3 months of age

Fatty acids (g/mg)	Median intake (n = 2,913)	Interquartile range (IQR)	Spearman's correlation coefficient between maternal intake of fatty acids and the child's serum fatty acid concent-ration at 3 months of age
Total fatty acids (g)	88.2	70.7–109.0	–
SFA (g)	37.8	29.3–47.9	–
Myristic (14:0) (mg)	4,007	3,008–5,408	0.14
Pentadecanoic (15:0) (mg)	518	377–710	0.43***
Palmitic (16:0) (mg)	17,884	14,200–22,538	0.14
Stearic (18:0) (mg)	9,059	7,092–11,516	0.30**
MUFA (g)	32.1	25.7–39.9	–
Palmitoleic (16:1 n-7) (mg)	1,078	849–1,360	−0.04
Palmitoleic (16:1 n-9) (mg)	256	197–330	−0.01
Oleic (18:1 n-9) (mg)	14,301	8,279–20,467	−0.06
Cis vaccenic (18:1 n-7) (mg)	830	495–1,156	–0.03
PUFA (g)	12.4	9.7–15.5	–
n-3 (g)	2.7	2.0–3.5	–
Alphalinolenic (18:3 n-3) (mg)	2,140	1,593–2,829	0.18*
Eicosapentaenoic (20:5 n-3) (mg)	74	38–125	0.26**
Docosahexaenoic (22:6 n-3) (mg)	214	117–356	0.09
n-6 (g)	9.4	7.4–11.7	–
Linoleic (18:2 n-6) (mg)	9,054	7,116–11,343	0.15
Arachidonic (20:4 n-6) (mg)	101	79–130	−0.01
Ratio of n-6:n-3	3.5	3.1–3.9	–
Conjugated linoleic (18:2) (mg)	154	113–208	0.43***

*** $p < 0.0001$; ** $p < 0.01$; * $p < 0.05$

eicosapentaenoic acid (EPA) from food during lactation correlated significantly with child's serum fatty acid concentrations at 3 months of age during exclusive breastfeeding (Table 1). Maternal fatty acid intake and consumption of their food sources correlated strongly with corresponding intake during pregnancy. Spearman's correlation coefficients for fatty acid or food intake during pregnancy versus lactation ranged between 0.47 and 0.78.

There were no statistically significant associations between maternal intake of fatty acids during lactation and the risk of preclinical or clinical type 1 diabetes (Table 3). The adjustment for the vocational education of the mother, genetic risk group, familial diabetes and exclusive duration of breastfeeding did not change the results.

There were some associations between maternal consumption of foods which are important sources of fatty acids during lactation and the endpoints (Table 4). After adjustment for potential confounders, maternal total consumption of red meat and meat products during lactation was associated with increased risk of preclinical [HR per 60 g increase in consumption of red meat and meat products was 1.19 (95 % CI 1.02–1.40), $p = 0.038$) and clinical type 1 diabetes [HR per 60 g increase in consumption of red meat and meat products was 1.27 (95 % CI 1.06–1.52), $p = 0.025$] (bold values in Table 4). To determine which component of meat and meat products accounts for the observed associations, we examined separately the consumption of unprocessed red meat (beef, pork, game, lamb and offals) and processed meat (meat products and sausages). After adjustment for potential confounders, there were no associations between consumption of unprocessed meat and endpoints, while maternal consumption of processed meat during lactation showed an association with increased risk for type 1 diabetes [HR per 34 g increase in consumption of processed meat was 1.23 (95 % CI 1.02–1.48), $p = 0.045$]. In addition to meat-associated food variables, maternal use of vegetable oils was associated with increased risk of preclinical type 1 diabetes [HR per 8 g increase in consumption of oils was 1.21 (95 % CI 1.03–1.41)].

Discussion

In the present birth cohort with genetic susceptibility to type 1 diabetes, maternal total consumption of red meat

Table 2 Maternal daily consumption of foods during lactation ($n = 2{,}913$) in 1998–2004

Food groups (g per day/frequency per week)	Median consumption during lactation	Interquartile range (IQR)
Milk and milk products (g)	669	417–930
Fresh milk (g)	403	154–643
Cheese (g)	49	26–82
Sour milk products (g)	126	59–218
Butter and butter-oil mixes (g)	9	4–19
High-fat margarine (g)	0	0–9
Low-fat margarine (g)	0	0–0.5
Oils (g)	10	6–16
Red meat and meat products (g)	107	79–142
Unprocessed meat[a] (g)	65	47–87
Processed meat[b] (g)	38	24–59
Poultry (g)	25	23–50
Fatty fish (times per week)	0.9	0.5–1.5
Lean fish (times per week)	0.5	0.2–0.9

[a] Food group of unprocessed red meat included beef, pork, game, lamb and offals

[b] Food group of processed meat included meat products and sausages

and meat products during lactation was associated with increased risk of both preclinical and clinical type 1 diabetes in the offspring. Separate analysis for unprocessed red meat did not show any association with the endpoints, while processed meat was associated with increased risk for clinical type 1 diabetes. Maternal consumption of vegetable oils during lactation was associated with increased risk of preclinical type 1 diabetes. No associations were observed between maternal intake of n-3 or other fatty acids and the risk of the two endpoints studied.

The strengths of the current study are the prospective cohort study design, the use of both preclinical and clinical type 1 diabetes as endpoints, and the relatively large number of endpoints. We observed expected associations between certain fatty acids in maternal diet and respective fatty acids in child's serum, suggesting that our measures of the maternal diet provide a reliable estimate.

There are, however, some limitations which imply that the present findings should be interpreted with caution. The major limitation of our study is the high number of tested associations. All together we tested over 70 associations including all fatty acid and food variables and two different endpoints. The level of statistical significance $p = 0.05$ connotes that with 20 tests, one test might be significant due to pure chance. Thus, it is possible that some of the observed associations are false-positive findings. Another limitation is that we did not take into account the child's own diet during follow-up period. Accordingly, the current results may not be attributable to the maternal intake during lactation alone. According to the validation study of the FFQ method applied in this survey, Pearson's correlation coefficients for intake of meat and oils were relatively low [26]. However, this may have decreased our ability to find associations rather than resulting in false findings. Maternal diet may also reflect some other lifestyle factors, which were not taken into account in the statistical models of the present study.

The present observation that maternal consumption of red meat and meat products during lactation was associated with both preclinical and clinical type 1 diabetes is the first prospective observation of this kind. In an earlier Sardinian case–control study, a trend toward a significant association between maternal meat consumption during pregnancy and lactation and higher risk of clinical type 1 diabetes ($p = 0.059$) was reported [29]. In a case–control study by Muntoni et al. [30], the child's own meat consumption, especially during the first 2 years of age, and also in later life was associated with type 1 diabetes in a dose–response manner. Also an ecological study in several countries showed a positive association between the national animal food intake, both dairy and meat, and type 1 diabetes incidence. In a nested case–control study within the DIPP cohort, we did not observe any association between the child's meat consumption and risk of beta-cell autoimmunity [31]. Neither did two prospective cohort studies show any association between maternal meat consumption during pregnancy and risk of preclinical or clinical type 1 diabetes [22, 32]. In the DIPP cohort, the biomarkers of milk or ruminant meat fat in the child's serum were associated with higher risk of autoimmunity [27]. Feskens et al. [33] recently discussed putative factors and mechanisms how meat consumption could increase the risk of both type 1 and type 2 diabetes, mentioning, e.g., SFA and trans fatty acids, dietary cholesterol, protein and amino acids, hemiiron, sodium, nitrites and nitrosoamines, and advanced glycation endproducts (AGE).

In the present study, separate analysis for unprocessed red meat did not show association with endpoints, while processed meat was associated with increased risk for clinical type 1 diabetes suggesting that industrial meat processing might result in harmful components, which transfer also to breast milk. Potential detrimental factors in processed meat could be nitrite and N-nitroso compounds [33], which have been observed to be associated with type 1 diabetes [34, 35]. However, maternal nitrate and nitrite intake was not reflected in the breast milk content [36]. Breast milk contains relatively high concentrations of nitrite and nitrate, and it has been implicated that breast milk nitrite and nitrate may be beneficial for the microbial colonization of gut in early life [37, 38]. The composition of intestinal microbiota may play a role in the development of type 1 diabetes [39].

Table 3 Risk of preclinical and clinical type 1 diabetes (hazard ratio, HR) in children with HLA-conferred susceptibility to type 1 diabetes associated with maternal fatty acid intake during lactation

Fatty acids[a] (g/mg)	Preclinical type 1 diabetes[b]				Type 1 diabetes			
	N total = 2,939 Cases, *n* = 172		*N* total = 2,939 Cases, *n* = 172		*N* total = 2,939 Cases, = 81		*N* total = 2,939 Cases, *n* = 81	
	Unadjusted HR (95 % CI)	*p* value[c]	Adjusted HR[d] (95 % CI)	*p* value[c]	Unadjusted HR (95 % CI)	*p* value[c]	Adjusted HR[d] (95 % CI)	*p* value[c]
Total fatty acids (g)	1.00 (0.85–1.17)	0.995	1.00 (0.85–1.17)	0.996	1.01 (0.81–1.26)	0.929	1.03 (0.83–1.28)	0.784
SFA (g)	0.93 (0.79–1.09)	0.346	0.94 (0.80–1.11)	0.471	0.94 (0.75–1.17)	0.559	0.97 (0.78–1.22)	0.823
Myristic (14:0) (mg)	0.89 (0.76–1.05)	0.153	0.91 (0.77–1.07)	0.243	0.91 (0.73–1.14)	0.428	0.95 (0.76–1.19)	0.657
Pentadecanoic (15:0) (mg)	0.89 (0.76–1.04)	0.147	0.91 (0.77–1.07)	0.233	0.92 (0.74–1.15)	0.450	0.95 (0.76–1.19)	0.659
Palmitic (16:0) (mg)	0.94 (0.80–1.11)	0.467	0.96 (0.81–1.13)	0.588	0.96 (0.73–1.20)	0.731	1.00 (0.80–1.25)	0.979
Stearic (18:0) (mg)	0.97 (0.83–1.14)	0.741	0.99 (0.84–1.17)	0.905	0.96 (0.77–1.20)	0.708	0.99 (0.79–1.25)	0.948
MUFA (g)	1.05 (0.90–1.22)	0.575	1.03 (0.89–1.21)	0.673	1.06 (0.86–1.31)	0.603	1.06 (0.86–1.31)	0.589
Palmitoleic (16:1 n-7) (mg)	0.93 (0.80–1.10)	0.397	0.93 (0.80–1.10)	0.408	0.99 (0.80–1.24)	0.943	1.01 (0.82–1.25)	0.919
Palmitoleic (16:1 n-9) (mg)	0.98 (0.83–1.14)	0.759	0.97 (0.83–1.14)	0.739	1.05 (0.85–1.30)	0.681	1.04 (0.85–1.27)	0.715
Oleic (18:1 n-9) (mg)	1.06 (0.91–1.24)	0.435	1.05 (0.90–1.23)	0.537	1.06 (0.85–1.31)	0.612	1.01 (0.81–1.25)	0.967
Cis vaccenic (18:1 n-7) (mg)	1.07 (0.91–1.24)	0.420	1.05 (0.90–1.23)	0.521	1.13 (0.91–1.39)	0.274	1.07 (0.87–1.32)	0.534
PUFA (g)	1.13 (0.98–1.32)	0.098	1.11 (0.95–1.29)	0.187	1.10 (0.90–1.35)	0.357	1.07 (0.87–1.31)	0.507
n-3 (g)	1.07 (0.92–1.24)	0.409	1.04 (0.89–1.22)	0.606	1.10 (0.90–1.36)	0.353	1.09 (0.89–1.34)	0.393
Alphalinolenic (18:3 n-3) (mg)	1.10 (0.95–1.28)	0.207	1.08 (0.93–1.26)	0.318	1.15 (0.94–1.40)	0.184	1.14 (0.93–1.39)	0.223
Eicosapentaenoic (20:5 n-3) (mg)	0.96 (0.82–1.13)	0.625	0.94 (0.79–1.11)	0.444	0.95 (0.75–1.19)	0.635	0.94 (0.74–1.20)	0.621
Docosahexaenoic (22:6 n-3) (mg)	0.97 (0.82–1.14)	0.695	0.94 (0.79–1.12)	0.488	0.95 (0.75–1.20)	0.665	0.95 (0.75–1.20)	0.658
n-6 (g)	1.14 (0.99–1.27)	0.070	1.12 (0.97–1.30)	0.130	1.09 (0.90–1.33)	0.401	1.06 (0.87–1.30)	0.583
Linoleic (18:2 n-6) (mg)	1.15 (1.00–1.32)	0.063	1.13 (0.97–1.30)	0.120	1.10 (0.90–1.33)	0.364	1.07 (0.87–1.30)	0.545
Arachidonic (20:4 n-6) (mg)	1.08 (0.93–1.26)	0.312	1.05 (0.91–1.22)	0.497	1.12 (0.91–1.36)	0.302	1.10 (0.91–1.32)	0.342
Gamma-linolenic (18:3 n-6) (mg)	0.88 (0.74–1.03)	0.113	0.88 (0.75–1.04)	0.132	0.94 (0.75–1.17)	0.579	0.96 (0.77–1.19)	0.705
Dihomo-γ-linolenic (20:3 n-6) (mg)	1.05 (0.90–1.22)	0.541	1.05 (0.90–1.21)	0.543	1.09 (0.90–1.32)	0.399	1.10 (0.93–1.31)	0.297
Ratio of n-6:n-3	1.05 (0.91–1.22)	0.477	1.05 (0.91–1.22)	0.496	0.94 (0.75–1.19)	0.625	0.91 (0.72–1.15)	0.431
Conjugated linoleic (18:2) (mg)	0.89 (0.75–1.04)	0.139	0.89 (0.76–1.05)	0.167	0.93 (0.74–1.16)	0.508	0.95 (0.76–1.18)	0.643

[a] Fatty acid variables used in statistical analyses were energy-adjusted by Willett's residual method. For fatty acid variables, hazard ratio (HR) describes change in risk, when the energy-adjusted fatty acid variable is changed to an extent corresponding to its standard deviation (SD)

[b] Testing positive for islet cell autoantibodies in combination with one or more of the autoantibodies against insulin, glutamic acid decarboxylase or tyrosine phosphatase-related islet antigen 2, or having clinical type 1 diabetes

[c] Likelihood ratio test was used to test whether the model with and without the food variables differed

[d] Model is adjusted for genetic risk group (0 = moderate), familial diabetes (0 = no), maternal vocational education (1 = none) and duration of exclusive breastfeeding (continuous)

Table 4 Risk of preclinical and clinical type 1 diabetes (hazard ratio, HR) in children with HLA-conferred susceptibility to type 1 diabetes associated with maternal consumption of foods which are sources of fatty acids during lactation

Food groups[a] (g per day/times per week)	Preclinical type 1 diabetes[b]				Type 1 diabetes			
	N total = 2,939 Cases, *n* = 172		*N* total = 2,939 Cases, *n* = 172		*N* total = 2,939 Cases, *n* = 81		*N* total = 2,939 Cases, *n* = 81	
	HR[c] (95 % CI)	*p* value[d]	Adjusted HR[e] (95 % CI)	*p* value[d]	HR[c] (95 % CI)	*p* value[d]	Adjusted HR[e] (95 % CI)	*p* value[d]
Milk and milk products (continuous) (g)	1.07 (0.89–1.28)	0.460	1.07 (0.89–1.29)	0.457	0.93 (0.72–1.20)	0.57	0.93 (0.71–1.21)	0.606
Cheese (continuous) (g)	0.89 (0.75–1.07)	0.210	0.84 (0.70–1.01)	0.053	0.94 (0.74–1.19)	0.582	0.87 (0.69–1.11)	0.246
Sour milk products (continuous) (g)	1.06 (0.91–1.24)	0.467	1.04 (0.89–1.22)	0.632	1.15 (0.95–1.39)	0.183	1.09 (0.89–1.33)	0.444
Fresh milk								
Lowest quarter: <154	1.16 (0.79–1.72)		1.18 (0.79–1.76)		1.66 (1.00–2.76)		1.62 (0.97–2.71)	
Intermediate half	1	0.333	1	0.300	1	0.134	1	0.188
Highest quarter: >643	1.33 (0.91–1.93)		1.35 (0.92–2.00)		1.04 (0.59–1.84)		1.12 (0.62–2.00)	
Butter and butter-oil mixes (g)								
Lowest quarter: <4	1.31 (0.90–1.91)		1.19 (0.80–1.76)		1.38 (0.80–2.37)		1.24 (0.71–2.15)	
Intermediate half	1	0.376	1	0.674	1	0.376	1	0.442
Highest quarter: >19	1.09 (0.73–1.62)		1.10 (0.74–1.65)		1.36 (0.80–2.31)		1.40 (0.82–2.39)	
High-fat margarine (g)	1.10 (0.95–1.28)	0.196	1.09 (0.94–1.26)	0.256	1		1.14 (0.95–1.37)	0.177
Users versus non-users	1.30 (0.95–1.79)	0.100	1.28 (0.92–1.77)	0.137	1.27 (0.82–1.96)	0.290	1.33 (0.85–2.07)	0.213
Low-fat margarine	0.90 (0.75–1.07)	0.217	0.90 (0.75–1.08)	0.234	1		0.87(0.67–1.14)	0.284
Users versus non-users	1.01 (0.72–1.42)	0.957	1.02 (0.72–1.44)	0.930	1.01 (0.62–1.61)	0.977	0.97 (0.60–1.57)	0.889
Oils (continuous) (g)	**1.23 (1.06–1.44)**	**0.008**	**1.21 (1.03–1.41)**	**0.023**	1.16 (0.95–1.42)	0.159	1.10 (0.89–1.37)	0.392
Red meat and meat products (continuous) (g)	**1.19 (1.01–1.40)**	**0.047**	**1.19 (1.02–1.40)**	**0.038**	**1.26 (1.04–1.53)**	**0.035**	**1.27 (1.06–1.52)**	**0.025**
Unprocessed meat[f]								
Lowest quarter: <47	1.20 (0.81–1.80)		1.12 (0.75–1.69)		1.55 (0.89–2.70)		1.40 (0.81–2.45)	
Intermediate half	1	0.212	1	0.321		0.131		0.259
Highest quarter: >87	1.40 (0.95–2.06)		1.36 (0.91–2.02)		1.60 (0.93–2.72)		1.50 (0.87–2.60)	
Processed meat[g] (continuous) (g)	1.11 (0.95–1.30)	0.196	1.12 (0.96–1.32)	0.155	1.21 (1.01–1.46)	0.058	**1.23 (1.02–1.48)**	**0.045**
Poultry (continuous) (g)	1.04 (0.90–1.21)	0.576	1.03 (0.88–1.20)	0.734	1.13 (0.96–1.33)	0.187	1.11 (0.94–1.32)	0.264
Fatty fish (times per week)								
Lowest quarter: <0.5	0.88 (0.61–1.26)		0.92 (0.63–1.33)		0.94 (0.59–1.52)		0.97 (0.59–1.58)	
Intermediate half		0.359	1	0.598		0.082	1	0.157
Highest quarter: >1.5	0.74 (0.49–1.13)		0.80 (0.52–1.23)		0.51 (0.27–0.97)		0.56 (0.29–1.07)	
Lean fish (times per week)								
Lowest quarter: <0.2	0.70 (0.50–1.00)		0.73 (0.51–1.04)		0.62 (0.38–1.01)		0.63 (0.38–1.03)	
Intermediate half		0.137	1	0.217		0.143	1	0.108

Table 4 continued

Food groups[a] (g per day/times per week)	Preclinical type 1 diabetes[b]				Type 1 diabetes			
	N total = 2,939 Cases, *n* = 172		*N* total = 2,939 Cases, *n* = 172		*N* total = 2,939 Cases, *n* = 81		*N* total = 2,939 Cases, *n* = 81	
	HR[c] (95 % CI)	*p* value[d]	Adjusted HR[e] (95 % CI)	*p* value[d]	HR[c] (95 % CI)	*p* value[d]	Adjusted HR[e] (95 % CI)	*p* value[d]
Highest quarter: >0.9	0.89 (0.58–1.37)		0.84 (0.54–1.31)		0.74 (0.40–1.35)		0.60 (0.32–1.14)	

[a] For continuous food variables, hazard ratio (HR) describes change in risk, when food variable is changed by an amount corresponding to its standard deviation (SD). For categorical food variables, reference category is intermediate half, and for dichotomized variables, reference category is non-users

[b] Testing positive for islet cell autoantibodies in combination with one or more of the autoantibodies against insulin, glutamic acid decarboxylase or tyrosine phosphatase-related islet antigen 2, or having clinical type 1 diabetes

[c] Model is adjusted for maternal energy intake

[d] Likelihood ratio test was used to test whether the model with and without the food variables differed

[e] Model is adjusted for maternal energy intake, genetic risk group (0 = moderate), familial diabetes (0 = no), maternal vocational education (1 = none) and duration of exclusive breast-feeding (continuous)

[f] Food group of unprocessed red meat included beef, pork, game, lamb and offals

[g] Food group of processed meat included meat products and sausages

One potential risk factor in meat could be proinflammatory AGE, which are found in heat-processed foods [40]. In a US study, high concentrations of AGE were observed in meat, fat and oils, when analyses was carried out using the enzyme-linked immunosorbent assay [40]. On the contrary, in a German AGE database, for which the AGE concentration have been measured by high-performance liquid chromatography, the AGE concentration for oils was very low [41]. AGEs have immunomodulative effects and both AGE and the receptor for AGE (RAGE) have been associated with increased risk for type 1 diabetes [42]. Maternal serum concentration of AGE correlated with newborn AGE status suggesting that AGEs could be transferred via placenta and also breast milk contained AGEs suggesting maternal transfer of AGE to breast milk [43].

In the current study, maternal use of vegetable oils during lactation was associated with higher risk of preclinical diabetes, which has not been observed before. We did not observe a consistent association with the fatty acid level (LA, ALA). However, there was a tendency toward a significant association between maternal LA intake during lactation and higher risk of preclinical type 1 diabetes (unadjusted $p = 0.063$). n-6 fatty acids may promote inflammation by inhibition of the synthesis of n-3 PUFA and acting as a precursor of proinflammatory eicosanoids [4].

We did not observe associations between maternal consumption of fish or intake of EPA or DHA during lactation and child's risk to develop type 1 diabetes. However, maternal consumption of fatty fish showed a trend for a protective effect against type 1 diabetes (unadjusted $p = 0.08$). Among children, there are prospective findings which suggest that n-3 fatty acids could protect the child from β-cell autoimmunity [6, 7], but not from progression of β-cell autoimmunity to clinical type 1 diabetes [44]. Maternal intake of n-3 or n-6 fatty acids or use of cod liver oil during pregnancy was not associated with autoimmunity or type 1 diabetes in two prospective studies [45, 46] nor in one case–control study [8].

The current results are the first prospective observations in which maternal meat, especially processed meat, and oil consumption during lactation were associated with higher risk of type 1 diabetes. However, taken into account the limitations of our study, the results do not give strong evidence and thus do not allow definitive conclusions. Maternal diet during lactation may also be a confounder when analyzing the associations between breastfeeding and risk of type 1 diabetes.

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Conflict of interest Sari Niinistö, Hanna-Mari Takkinen, Liisa Uusitalo, Jenna Rautanen, Noora Vainio, Suvi Ahonen, Jaakko Nevalainen, Michael G Kenward, Mirka Lumia, Olli Simell, Riitta Veijola, Jorma Ilonen, Mikael Knip and Suvi M. Virtanen declare that they have no conflict of interest.

Ethical standard The study was approved by the Research Ethics Committee of the Universities of Tampere and Oulu.

Human and animal rights disclosure All procedures followed were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2008.

Informed consent disclosure Informed consent was obtained from all patients for being included in the study.

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