

ORIGINAL ARTICLE

EPIDEMIOLOGY AND GENETICS

# Breast milk fatty acid composition has a long-term effect on the risk of asthma, eczema, and sensitization

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**To cite this article:** van Elten TM, van Rossem L, Wijga AH, Brunekreef B, de Jongste JC, Koppelman GH, Smit HA. Breast milk fatty acid composition has a long-term effect on the risk of asthma, eczema, and sensitization. *Allergy* 2015; **70**: 1468–1476.

## Keywords

Asthma; breast milk fatty acids; childhood; eczema; sensitization.

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Accepted for publication 21 July 2015

DOI:10.1111/all.12703

Edited by: Antonella Muraro

## Abstract

**Background:** Levels of n-3 polyunsaturated fatty acids (PUFAs) and n-6 PUFAs in breast milk are associated with the development of allergic diseases up to school age. However, it is unknown whether this relationship persists when the child becomes older. We therefore studied the association between levels of n-3 PUFAs and n-6 PUFAs in breast milk of allergic- and nonallergic mothers and asthma, eczema and sensitization up to the age of 14 years.

**Methods:** The study was nested in the ongoing PIAMA birth cohort. At the child's age of 3 months, 276 mothers provided a breast milk sample. Asthma ( $N$  total = 269) and eczema ( $N$  total = 274) were self-reported up to the child's age of 14 years. Specific serum IgE levels were measured at the ages of 4, 8 and 12 years ( $N$  total = 216). Generalized estimating equations analyses were used to take account of repeated observations.

**Results:** Asthma up to the age of 14 years is less prevalent in children of allergic mothers receiving breast milk with higher levels of n-3 long chain polyunsaturated (LCP) fatty acids (OR 0.50; 95% CI 0.31–0.79), and more prevalent in children of nonallergic mothers receiving breast milk with higher levels of n-6LCP (OR 1.86; 95% CI 1.14–3.03). Weaker associations in similar direction were observed for eczema and sensitization. Direction of associations were consistent and of similar magnitude throughout childhood.

**Conclusion:** The association between breast milk fatty acid composition and asthma, eczema and sensitization persists up to the age of 14 years in children of both allergic and nonallergic mothers.

Early life fatty acid intake is one of the factors that plays a role in the development of allergic diseases in early childhood. However, it is unknown if this effect persists when the child becomes older. The influence of fatty acids on the

risk of developing allergic diseases was suggested by the rising prevalence of these diseases over the last decades (1, 2) combined with the increased consumption of linoleic acid and the decreased consumption of oily fish (3). The latter is

## Abbreviations

AA, Arachidonic acid (C20:4n-6); ALA,  $\alpha$ -linoleic acid (C18:3n-3); CI, Confidence interval; DHA, Docosahexaenoic acid (C22:6n-3); EPA, Eicosapentaenoic acid (C20:5n-3); IgE, Immunoglobulin E; LA, Linoleic acid (C18:2n-6); n-3 PUFAs, n-3 polyunsaturated fatty acids; n-3LCP/n-6LCP, Ratio between n-3 and n-6 long chain polyunsaturated fatty acids; n-3LCP, Total n-3 long chain polyunsaturated fatty acids ( $C \geq 20$ ); n-6 PUFAs, n-6 polyunsaturated fatty acids; n-6LCP, Total n-6 long chain polyunsaturated fatty acids ( $C \geq 20$ ); OR, Odds ratio; PIAMA, Prevention and Incidence of Asthma and Mite Allergy; PUFAs, Polyunsaturated fatty acids; RAST, Radioallergosorbent test.

an important source of n-3 long chain polyunsaturated fatty acids (LCP). In particular, the n-6 polyunsaturated fatty acid (PUFA) linoleic acid, n-3 PUFA  $\alpha$ -linoleic acid and their long chain derivatives arachidonic acid (AA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA) seem to be associated with allergic diseases. N-3 PUFAs appear to have a protective effect, while a high intake of n-6 PUFAs seemed to increase the risk of allergic diseases (4–8).

Because allergic diseases often develop in early childhood (8), infants are an important target population for primary allergy prevention. Therefore, the association between fatty acid composition in mothers' breast milk and the development of allergic diseases has been studied extensively (9–18). As this relationship has generally been studied up until preschool age (9–14, 16–18), it is unknown whether observed associations between breast milk fatty acids and allergic diseases persist when the child becomes older.

Previously, we reported the associations between level of breast milk n-3 PUFAs and n-6 PUFAs and allergic symptoms at the age of 1 and 4 years. In this study, we observed that the risk to develop allergic symptoms in children of allergic mothers, but not the risk of sensitization, was modified by the intake of n-3LCP throughout breast milk (10). In the current study, we extend our research by examining the association between the level of n-3 PUFAs and n-6 PUFAs in mothers' breast milk and the development of asthma, eczema and sensitization in their children up to the age of 14 years, stratified on maternal allergy.

## Methods

### Design and study population

The current study was embedded in the Prevention and Incidence of Asthma and Mite Allergy (PIAMA) birth cohort (19, 20). Pregnant women were recruited from the general population during their first antenatal visit. They filled out a validated short screening questionnaire to classify them as allergic or nonallergic ('yes' when reported (a history of) asthma, current hay fever, current allergy for pets, current allergy for house dust or house dust mite, otherwise 'no') (21). All children ( $N = 3963$ ) were born in 1996/1997 and are followed up until present. Questionnaires were completed by the parents during pregnancy, at the child's age of 3 months, annually from 1 year up until 8, 11 and 14 years. Additionally, in all children of allergic mothers ( $N = 1327$ ) and in a random sample of children of nonallergic mothers ( $N = 663$ ), a home visit was performed around the age of 3 months. Extensive clinical examinations were performed at the child's age of 4, 8 and 12 years. The study protocol was approved by the Medical Ethical Committees of the participating institutions, and all participants gave written informed consent.

Eligible for the current study were all children from mothers who had provided a breast milk sample. Breast milk was collected during the home visit to 1860 families (93% of those selected for home visit). At that time, 661 of the visited mothers were still breastfeeding, and 276 of them were willing and able to produce a breast milk sample by manual

expression or breast pump. Of the 276 children who had a breast milk sample, 269 children had data on asthma, 274 children on eczema and 216 children on sensitization.

### Breast milk fatty acids

Breast milk samples were collected around 3 months postpartum (mean 15.1 weeks in allergic mothers [SD: 4 weeks]; mean 15.7 weeks in nonallergic mothers [SD: 4.3 weeks]). All samples were put into tubes that contained 2  $\mu$ l of butylated hydroxyl toluene solution (500  $\mu$ g/ml) to prevent oxidation and stored at  $-70^{\circ}\text{C}$ . Thereafter, gas-liquid chromatography was used to determine the fatty acid composition of the breast milk, as described by Foreman-van Drongelen (22). Polyunsaturated, monounsaturated and saturated fatty acids were measured as weight percentage (wt%) of the total breast milk fatty acid composition. Details about the collection of breast milk and analysis have been reported previously (23).

Based on previous studies, we were interested in the following fatty acids: linoleic acid (LA; C18:2n-6), arachidonic acid (AA; C20:4n-6), total n-6 long chain ( $C \geq 20$ ) polyunsaturated fatty acids (n-6LCP),  $\alpha$ -linoleic acid (ALA; C18:3n-3), eicosapentaenoic acid (EPA; C20:5n-3), docosahexaenoic acid (DHA; C22:6n-3), total n-3 long chain ( $C \geq 20$ ) polyunsaturated fatty acids (n-3LCP), and the ratio between n-3 and n-6 long chain ( $C \geq 20$ ) polyunsaturated fatty acids (n3-LCP/n-6LCP).

### Asthma, eczema and sensitization

Both outcome variables asthma and eczema were based on parents' answers to questions adapted from the International Study on Asthma and Allergy in Children (ISAAC) questionnaire (24). Asthma was defined as having at least two of the following criteria: doctor's diagnosis of asthma ever, wheezing during the last 12 months and use of asthma medications prescribed by a doctor during the last 12 months. Data regarding asthma were available annually at child's age of 3 years up until 8, 11 and 14 years.

Eczema was defined as an itchy rash that was coming and going during the last 12 months (at folds of the elbows, or back of the knee, or front of the ankles, or in the neck, or around eyes and ears) and a doctor's diagnosis of eczema ever. Data regarding eczema were available at child's age of 3 months, annually from 1 year up until 8, 11 and 14 years.

At child's age of 4, 8 and 12 years, a blood sample was taken to measure specific IgE at Sanquin Research Amsterdam using RASTs. Sensitization was defined as specific IgE higher than 0.35 IU/ml to house dust mite, cat, grass pollen, birch pollen, egg or milk.

### Fixed covariates

Information about covariates was obtained from questionnaires. At the start of the study, data were gathered about the presence of older siblings of the child (yes/no; number), maternal age at delivery (years), maternal smoking during at

least the first 4 weeks of pregnancy (yes/no), maternal and paternal education (highest education completed, categorized into low/middle/high education level), maternal BMI before pregnancy (weight and height, kg/m<sup>2</sup> is calculated), gestational weight gain of the mother (in kg, categorized into <10 kg/≥10 kg), duration of any breastfeeding (in weeks, categorized into <16 weeks/≥16 weeks), child's gender (boy/girl), parental allergy based on a validated short screening questionnaire (21) as described earlier, having an allergic family member ('yes' when having an allergic mother, allergic father or ≥1 allergic sibling(s), otherwise 'no'), birthweight of the child (grams) and born by Caesarean section (yes/no). From child's age of 3 months up until 1 year, data were gathered about the introduction of fruit(juice), vegetables, milk(products), wheat products, meat, fish and eggs, categorized into 'food/beverage <4 months of age'/'food/beverage ≥4 months of age'.

#### Time varying covariates

Data about household smoking (yes/no), BMI of the child (weight and height, kg/m<sup>2</sup> and Z-scores according to a national reference were calculated) (25) and pet exposure ('yes' when exposed to cat, dog, rodent or bird at home, otherwise 'no') were gathered at child's age of 3 months, 1 year up until 8, 11 and 14 years. Smoking by the child ('daily' when at least once per day, 'occasional' when not every day but at least once per week or once per month or less than once a month or sometimes, 'former' when quit smoking, and 'never' when never smoked) was asked directly to the child via a questionnaire at the age of 11 and 14 years.

#### Statistical analysis

Statistical analyses were performed using SAS, version 9.2 (SAS Institute Inc., Cary, NC, USA). Because associations between fatty acids and asthma, eczema and sensitization may differ in children of allergic mothers compared with children of nonallergic mothers, we stratified on maternal allergy. Associations up to the age of 14 years were analysed by generalized estimating equations (GEE), using an exchangeable correlation structure to account for correlations between repeated observations in the same individual at the different points in time. Breast milk fatty acids were analysed as continuous exposure variables. To assess whether associations were consistent throughout childhood, breast milk fatty acids were added to the model as continuous variables, age was added to the model as categorical variable, and an interaction term between breast milk fatty acids and age was used to obtain age-specific estimates. We report odds ratios (ORs) and 95% confidence intervals (95% CI) per interquartile range increase in levels of breast milk fatty acid.

All fixed and time varying covariates as mentioned earlier were selected based on literature. Those covariates that were associated with both exposure and outcome variables in our database were added to the statistical model one at the time

to estimate adjusted ORs. Comparing these adjusted ORs to the crude ORs, it was concluded that associations were not affected by any of these covariates (difference in crude vs adjusted ORs <10%). Therefore, only crude ORs are shown.

The number of mothers smoking during pregnancy was too small to add as covariate in the model, as there were hardly any children with asthma of smoking nonallergic mothers, as well as the number of children smoking at the age of 11 (none) and 14 years (*N* = 10). Therefore, we checked the robustness of the association by restricting the sample to mothers who did not smoke during pregnancy and to nonsmoking children.

## Results

### General characteristics

The study population consisted of 274 children, 61% (*N* = 167) of allergic mothers and 39% (*N* = 107) of nonallergic mothers. Characteristics of the study population were similar for allergic and nonallergic mothers, with largest differences in maternal smoking during pregnancy (12.1% vs 7.6%, respectively; *P*-value = 0.234) and maternal education level (low educated 16.0% vs 11.4% and high educated 48.5% vs 41.9%, respectively; *P*-value = 0.175) (Table 1). No difference was seen in the fatty acid composition of breast milk of allergic mothers compared with nonallergic mothers, as reported in our previous study (23). Figure 1 shows the age-specific prevalence for asthma, eczema and sensitization.

### Children of allergic mothers

Children of allergic mothers receiving breast milk with a higher amount of n-3LCP were less likely to have asthma (OR 0.50; 95% CI 0.31–0.79), which was mainly attributable to DHA (OR 0.48; 95% CI 0.27–0.83) (Table 2). Children receiving breast milk with a higher n-3LCP/n-6LCP ratio were less likely to have asthma (OR 0.55; 95% CI: 0.33–0.92), which is driven by n-3LCP rather than by n-6LCP (OR 1.00; 95% CI 0.57–1.78). Although nonsignificant, children receiving breast milk with a higher amount of n-3LCP seem less likely to be sensitized (OR 0.77; 95% CI 0.57–1.04). No strong associations were seen between breast milk fatty acid intake and eczema.

### Children of nonallergic mothers

Children of nonallergic mothers receiving a higher amount of breast milk n-6LCP were more likely to have asthma (OR 1.86; 95% CI 1.14–3.03), which was mainly attributable to arachidonic acid (OR 2.12; 95% CI 1.22–3.70) (Table 3). No association was seen between total n-3LCP and asthma (OR 1.13; 95% CI 0.94–1.36). Children receiving a higher amount of n-3LCP were less likely to have eczema (OR 0.89; 95% CI 0.74–1.07), while children receiving a higher amount of arachidonic acid were more likely to have eczema (OR 1.37; 95% CI 1.03–1.81). This was also reflected in the association

**Table 1** Characteristics of the study population and fatty acid composition of the mothers' breast milk stratified by maternal allergy (%; N unless otherwise indicated)

	Allergic mothers (N = 167)	Nonallergic mothers (N = 107)
<b>Maternal characteristics</b>		
Maternal age at delivery (years; mean; SD)	31.1 (3.6)	30.8 (4.1)
Days after delivery milk sample was collected (mean; SD)	105.8 (27.9)	110.2 (29.8)
First child	53.3 (89)	53.3 (57)
Maternal smoking during pregnancy	12.1 (20)	7.6 (8)
Education level		
Low education level	16.0 (26)	11.4 (12)
Middle education level	35.6 (58)	46.7 (49)
High education level	48.5 (79)	41.9 (44)
<b>Paternal characteristics</b>		
Paternal allergy	32.9 (55)	29.0 (31)
Education level		
Low education level	15.4 (25)	20.2 (21)
Middle education level	30.3 (49)	34.6 (36)
High education level	54.3 (88)	45.2 (47)
<b>Characteristics of the children</b>		
Gender (% boys)	51.5 (86)	51.4 (55)
Birthweight (grams; mean; SD)	3532 (474)	3559 (522)
Smoking at the age of 14 years (% yes)	4.8 (8)	1.9 (2)
<b>Fatty acid composition of the breast milk (median wt%; IQR)</b>		
LA (C18:2n-6)	14.87 (5.82)	14.61 (4.20)
AA (C20:4n-6)	0.36 (0.12)	0.39 (0.11)
ALA (C18:3n-3)	0.92 (0.44)	0.97 (0.35)
EPA (C20:5n-3)	0.04 (0.03)	0.05 (0.03)
DHA (C22:6n-3)	0.17 (0.11)	0.16 (0.10)
Total n-6LCP	1.09 (0.34)	1.15 (0.29)
Total n-3LCP	0.48 (0.19)	0.49 (0.19)
n3-LCP/n-6LCP	0.44 (0.21)	0.43 (0.19)

between the ratio n-3LCP/n-6LCP and eczema (OR 0.82; 95% CI 0.65–1.03). Children receiving a higher amount of linoleic acid (n-6) in breast milk were more likely to be sensitized (OR 1.64; 95% CI 1.05–2.56).

#### Age-specific estimates

The direction of the associations between breast milk fatty acids and asthma, eczema and sensitization in children of allergic and nonallergic mothers was consistent at all ages (Figure 2, results only shown for the association between DHA and the ratio n-3LCP/n-6LCP and asthma).

#### Sensitivity analysis

Estimates hardly changed when restricting the analyses to children of mothers who did not smoke during pregnancy, as well as to nonsmoking children (results not shown).

## Discussion

We showed that the association between fatty acid intake during early life and asthma, eczema and sensitization persists up to the age of 14 years. Children of allergic mothers receiving breast milk with a higher amount of n-3LCP were less likely to have asthma, while children of nonallergic mothers receiving breast milk with a higher amount of n-6LCP were more likely to have asthma. Similar tendencies were seen in the associations between breast milk fatty acids and eczema and sensitization. The effects of specific fatty acids differed between children of allergic and nonallergic mothers, especially for asthma.

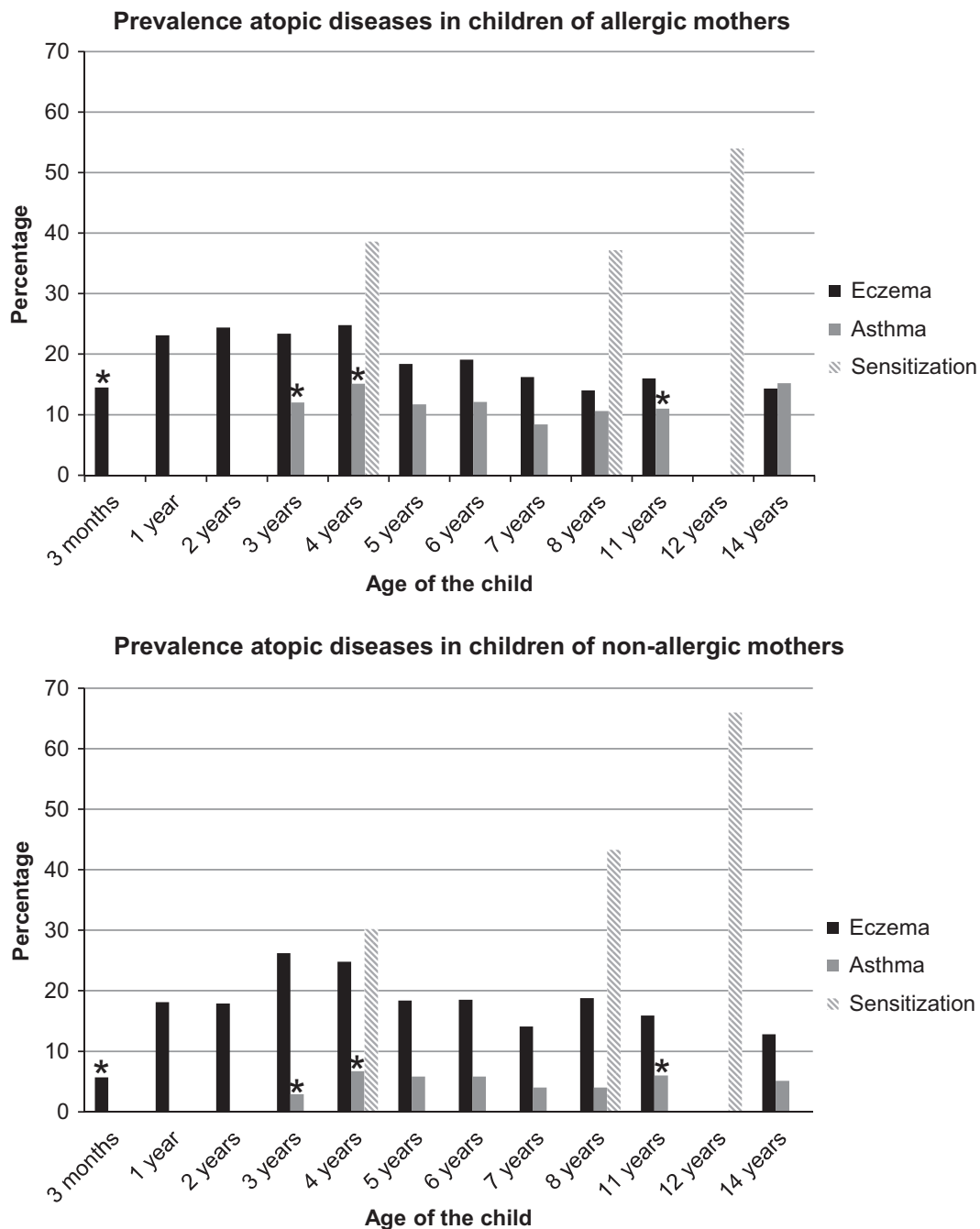
#### Strengths and limitations

No previous studies examined the association between breast milk fatty acids and asthma, eczema and sensitization up to the age of 14 years, taking into account multiple repeated measurements of allergic diseases and covariates. Using GEE analysis, full use was made of these repeated measurements, taking into account correlations between repeated observations in the same individual at different points in time.

We collected breast milk samples once when the child was about 3 months old. Evidence suggest that the percentage levels of DHA and arachidonic acid decrease over time (26, 27). This could have led to bias if mothers who collected breast milk at a later point in time, have children with a lower risk on allergic outcomes. This is unlikely, as we found no correlation between breast milk fatty acids and time since delivery when the samples were obtained (10). Most of our breast milk samples (86%) were collected around 3 months postpartum or later. Breast milk collection was not standardized regarding timing of the sampling (e.g. part of the day). The way in which the mother had produced the milk sample (by manual expression or using a breast pump, and before or after feeding the child) and the time of the day this was done did not influence the concentration of individual fatty acids (23). If misclassification occurred, this misclassification is unlikely to be differentially associated with the risk on allergic outcomes.

One study (224 children, follow-up 7 years) (15) found that the association between the fatty acid composition of colostrum and eczema was modified by gender of the child. This effect became weaker studying 3 months expressed breast milk. As the breast milk in our study was collected around 3 months postpartum, effect modification by gender of the child is unlikely. In addition, to exclude any effect of gender, we added gender as covariate into the model as our sample size was too small to stratify on maternal allergy as well as on gender of the child. The associations between breast milk fatty acids and asthma, eczema and sensitization hardly changed (difference crude vs adjusted ORs <5%; results not shown).

Asthma and eczema were both self-reported conditions. Both outcome variables were based on the standardized and worldwide-used ISAAC questionnaire (24), and the definition of asthma is in line with the definition



**Figure 1** Age-specific prevalence of asthma, eczema and sensitization throughout childhood in children of allergic and nonallergic mothers. \* $P < 0.05$ .

accomplished by the European MeDALL consortium (28). Although self-reporting of asthma and eczema could lead to misclassification, it is very unlikely that reporting error is related to breast milk fatty acids and therefore led to biased results.

The population selected for this study consists of higher educated mothers and more allergic mothers compared with

the PIAMA cohort. As participants were recruited without any knowledge of either exposure or disease status, selection bias is unlikely. In addition, the proportion of sensitized children of nonallergic mothers at the age of 12 years old is high compared with the proportion of sensitized children of allergic mothers. We cannot think of any specific selective mechanism that may have occurred resulting in biased asso-



**Table 2** Odds ratio (95% CI) reflecting the overall association up to the age of 12 years (sensitization) and 14 years (asthma/eczema) between breast milk fatty acid composition and allergy in children of allergic mothers†

FA (wt%)	Asthma (N = 162)		Eczema (N = 167)		Sensitization (N = 130)	
	OR	95% CI	OR	95% CI	OR	95% CI
LA (C18:2n-6)	1.47	0.86, 2.51	1.15	0.84, 1.58	0.85	0.56, 1.30
AA (C20:4n-6)	0.76	0.44, 1.32	0.88	0.64, 1.21	0.97	0.62, 1.51
ALA (C18:3n-3)	1.02	0.68, 1.53	0.81	0.58, 1.12	0.80	0.55, 1.16
EPA (C20:5n-3)	0.79	0.59, 1.05	0.90	0.69, 1.17	0.72*	0.55, 0.94
DHA (C22:6n-3)	0.48**	0.27, 0.83	0.90	0.67, 1.22	0.80	0.61, 1.04
Total n-6LCP	1.00	0.57, 1.78	1.24	0.90, 1.71	0.87	0.56, 1.33
Total n-3LCP	0.50**	0.31, 0.79	0.83	0.60, 1.15	0.77	0.57, 1.04
n-3LCP/n-6LCP	0.55*	0.33, 0.92	0.78	0.54, 1.13	0.91	0.69, 1.20

†Odds ratios were estimated for an interquartile range increase in breast milk fatty acid composition (wt%). An exchangeable correlation structure was used to account for correlations between repeated observations in the same individual.

\* $P < 0.05$ .

\*\* $P < 0.01$ .

**Table 3** Odds ratio (95% CI) reflecting the overall association up to the age of 12 years (sensitization) and 14 years (asthma/eczema) between breast milk fatty acid composition and allergy in children of nonallergic mothers†

FA (wt%)	Asthma (N = 107)		Eczema (N = 107)		Sensitization (N = 86)	
	OR	95% CI	OR	95% CI	OR	95% CI
LA (C18:2n-6)	0.74	0.27, 2.01	1.16	0.79, 1.71	1.64*	1.05, 2.56
AA (C20:4n-6)	2.12**	1.22, 3.70	1.37*	1.03, 1.81	1.01	0.66, 1.54
ALA (C18:3n-3)	1.03	0.41, 2.60	1.02	0.77, 1.35	1.26	0.83, 1.91
EPA (C20:5n-3)	1.04	0.80, 1.34	0.86	0.68, 1.08	1.17	0.95, 1.44
DHA (C22:6n-3)	1.10	0.95, 1.27	0.93	0.79, 1.09	1.06	0.89, 1.27
Total n-6LCP	1.86*	1.14, 3.03	1.11	0.83, 1.48	0.86	0.59, 1.24
Total n-3LCP	1.13	0.94, 1.36	0.89	0.74, 1.07	1.06	0.85, 1.32
n-3LCP/n-6LCP	1.01	0.82, 1.24	0.82	0.65, 1.03	1.12	0.92, 1.36

†Odds ratios were estimated for an interquartile range increase in breast milk fatty acid composition (wt%). An exchangeable correlation structure was used to account for correlations between repeated observations in the same individual.

\* $P < 0.05$ .

\*\* $P < 0.01$ .

ciations due to a relative high prevalence of sensitized children.

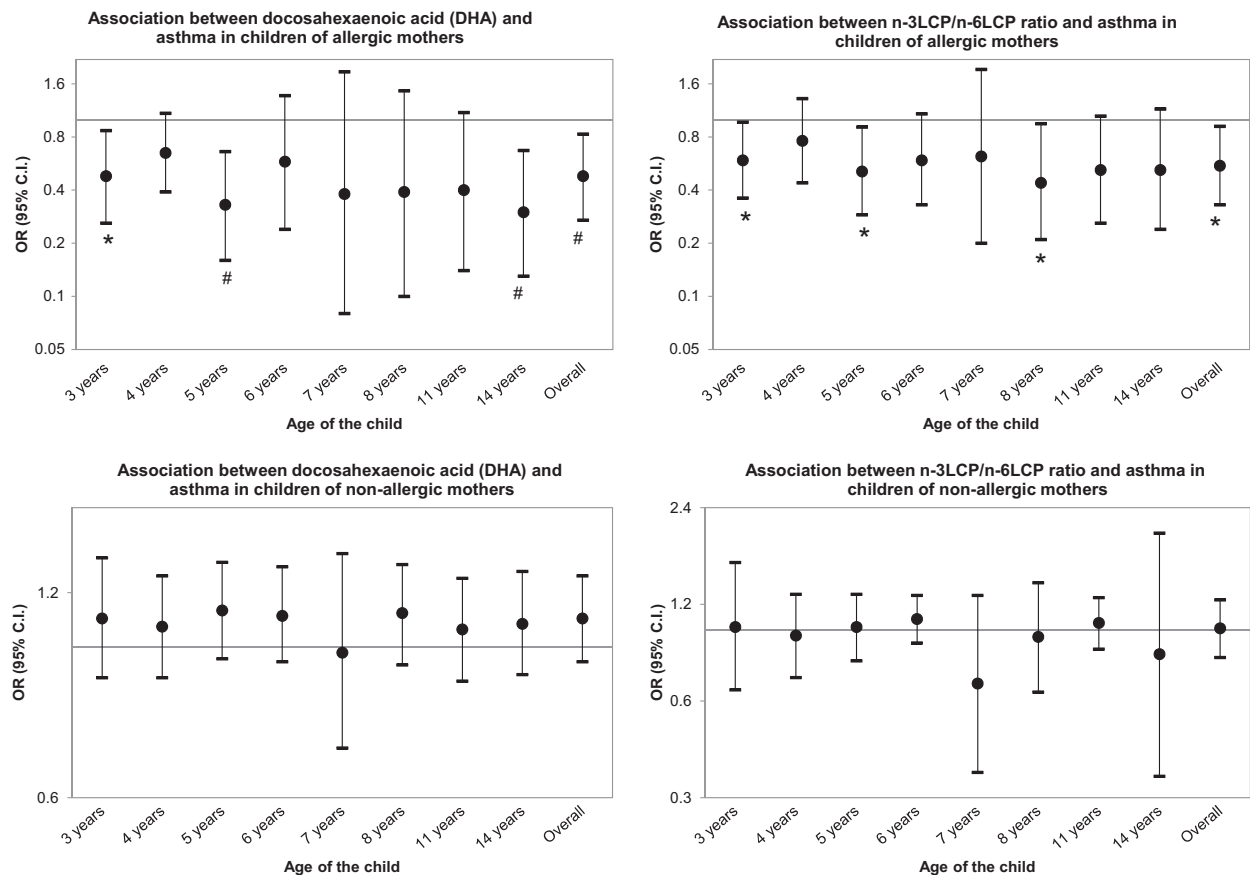
### Underlying mechanism

Predisposition to allergic diseases may result from insufficiently balanced T helper type 1 cell (Th1) and type 2 cell (Th2) pathways (5). A diet rich in n-6 fatty acids causes rise of eicosanoids such as prostaglandin (PGE<sub>2</sub>) formed by the action of cyclooxygenase on arachidonic acid (3, 5). This may promote interleukin-4 to increase the number of B-lymphocytes producing IgE (3). N-3 PUFA potentially influence the T helper cell balance via inhibiting interleukin-13 production, which is related to atopy via IgE synthesis in B-lymphocytes and Th2 differentiation in T cells (5). As linoleic acid and  $\alpha$ -linoleic acid are competitive for the same enzyme system for desaturation–elongation, consuming a diet rich in n-6 fatty acids may affect the n-3LCP/n-6LCP ratio, leading to predominance of arachidonic acid in tissues, which pro-

motes IgE production as described above (5). A study of D'Vaz et al. (29) confirms that post-natal fish oil supplementation up to 6 months of age decreased allergen-specific Th2 responses and increased polyclonal Th1 responses. The mechanism by which early fatty acid intake may influence later life allergy may be via programming. As differences in immune function associated with allergic diseases are already present at birth (30, 31) and immune function is programmed in early life and, a favourable n-3LCP/n-6LCP ratio during early development seems most likely to influence allergy development.

### Comparison with other studies

No previous studies examined the association between breast milk fatty acids and asthma, eczema and sensitization up to the age of 14 years. Therefore, we compare our results to the results of studies that examined the same association at different child's ages. The associations found in our study are



**Figure 2** Overall and age-specific associations between the breast milk fatty acids DHA and the ratio n-3LCP/n-6LCP and asthma in children of allergic and nonallergic mothers. Odds ratios were calcu-

lated for an interquartile range increase in breast milk fatty acid composition (wt%). \*  $P < 0.05$ , #  $P < 0.01$ .

in line with results of a number of previous studies (overview table available upon request) (9, 11–14, 16, 18).

In contrast to our results, Lowe et al. (15) (224 children, follow-up 7 years) found evidence that high levels of total n-3 PUFAs in breast milk were associated with increased risk of nonatopic eczema within the first 2 years of life compared with healthy children. To compare, we composed the variable nonatopic eczema, defined as reported eczema but no sensitization at the age of 4 years. We found no relation between nonatopic eczema and total n-3 PUFA (n-3LCP: OR 0.91 [0.61–1.36]; ALA: OR 1.16 [0.68–1.98]; nonstratified). Unlike the study of Lowe et al., we were not able to identify nonatopic eczema at the age of 2 years. In addition, Lowe et al. found no associations between the fatty acids profile in breast milk and the risk of childhood asthma at 6 or 7 years of age. Results were not stratified on allergy of the mother. To compare, we analysed the relation between breast milk fatty acids and asthma at the age of 7 years without stratification (DHA: OR 0.71 [0.35–1.45]; n-3LCP: OR 0.70 [0.41–1.21]; n-6LCP: OR 1.09 [0.56–2.14]; nonstratified) and found comparable associations. Reichardt et al. (17) (78 children pairs, follow-up 1 year) found no association

between breast milk fatty acids and atopic eczema maybe because colostrum was studied instead of mature breast milk.

#### Children of mothers with allergy and children of mothers without allergy

Why would the risk to develop asthma and possibly eczema and sensitization in children of allergic and nonallergic mothers be associated with different fatty acid profiles? First, there could be a difference in the optimal fatty acid composition for children with a genetic predisposition to develop asthma compared with low-risk children. Indeed, asthma at this age is a heterogeneous disorder and atopy is only one of the phenotypes in the pathogenesis. Another explanation could be that components in breast milk of allergic and nonallergic mothers we did not measure interacted with fatty acids, leading to different patterns of asthma development in their offspring. Some studies suggest a difference in the association between maternal breast milk fatty acids and allergic diseases in high-risk children compared with a low-risk population (12, 14).

## Implications and conclusion

We showed a long-term effect of breast milk fatty acid composition on the risk of asthma, and probably also eczema and sensitization. For the purpose of primary allergy prevention, dietary advice regarding fatty acid intake during pregnancy and lactation may play an important role, as the ratio of n-6 to n-3 PUFAs in diet has increased in most Western countries (32). A large body of literature examines the effects of maternal or early post-natal supplementation with n-3 PUFA to prevent allergic diseases (33–35). Despite of inconsistent results and the differential effects for maternal and child supplementation, the majority of studies shows a protective effect (33), which offers possibilities for prevention. Randomized controlled trials are needed to elucidate the differential effects of maternal allergy on the associations between breast milk fatty acid composition and childhood asthma, eczema and sensitization, and to define the optimal diet and target population on which public health recommendations can be based. In conclusion, breast milk fatty acid composition influences the risk of asthma, eczema and sensitization not only in early life, but also up to the age of 14 years.

## Acknowledgments

The PIAMA study was supported by the Netherlands Organization for Health Research and Development; The

Netherlands Organization for Scientific Research; The Netherlands Lung Fund; The Netherlands Ministry of Spatial Planning, Housing, and the Environment; and The Netherlands Ministry of Health, Welfare, and Sport.

## Conflicts of interest

The authors declare that they have no conflicts of interest.

## Authors contributions

T.M.E. participated in design of this study, data analysis, interpretation of results and drafted the manuscript; L.R. participated in design of this study, interpretation of results and revised the manuscript; A.H.W. contributed to the conception and design, acquisition of data, interpretation of results and revised the manuscript; B.B. contributed to the conception and design of the PIAMA study, acquisition of data, revised the manuscript; J.C.J. and G.H.K. contributed to the conception and design of the PIAMA study, acquisition of data and revised the manuscript; H.A.S. contributed to the conception and design of the PIAMA study, interpretation of results and revised the manuscript. All authors approved the final version of the manuscript.

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