

**Maternal fat consumption during pregnancy and risk of wheeze and eczema in Japanese infants aged 16-24 months: The Osaka Maternal and Child Health Study**

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## ABSTRACT

**Background:** One factor capable of modulating antenatal immune responses is diet. The present prospective study examined the association between maternal intake of specific types of fatty acids, cholesterol, fish, and meat during pregnancy and the risk of wheeze and eczema in the offspring.

**Methods:** Subjects were 763 mother-child pairs. Data on maternal intake during pregnancy were assessed with a diet history questionnaire. Data on wheeze and eczema based on criteria of the International Study of Asthma and Allergies in Childhood were obtained from our questionnaire completed by mothers 16-24 months postpartum.

**Results:** Higher maternal intake of  $\alpha$ -linolenic acid and docosahexaenoic acid during pregnancy was independently associated with a reduced risk of wheeze, but not eczema, in the offspring (adjusted ORs between extreme quartiles [95% CIs] = 0.52 [0.28–0.97] and 0.37 [0.15–0.91], respectively). Higher maternal intake of n-6 polyunsaturated fatty acids and linoleic acid during pregnancy was independently related to an increased risk of infantile eczema, but not wheeze (adjusted ORs [95% CIs] = 2.25 [1.13–4.54] and 2.11 [1.06–4.26], respectively). No significant exposure-response relationships were observed between maternal consumption of total fat, saturated fatty acids, monounsaturated fatty acids, eicosapentaenoic acid, arachidonic acid, cholesterol, meat, and fish and the ratio of n-3 to n-6 polyunsaturated fatty acid consumption during pregnancy and infantile wheeze or eczema.

**Conclusions:** Maternal intake of  $\alpha$ -linolenic acid and docosahexaenoic acid during pregnancy may be preventive against infantile wheeze. Maternal intake of n-6 polyunsaturated fatty acids, especially linoleic acid, during pregnancy may increase the risk of childhood eczema.

## INTRODUCTION

One of the factors capable of modulating antenatal immune responses is diet. Especially, n-3 and n-6 polyunsaturated fatty acids can influence the Th1 and Th2 balance.<sup>1</sup> A case-control study of US schoolchildren showed that maternal oily fish consumption at least monthly versus no consumption during pregnancy was significantly related to a decreased risk of early persistent asthma in the children.<sup>2</sup> A significant preventive relationship was observed between maternal fish intake during pregnancy and the risk of atopic wheeze in the offspring at 6 years of age in a Spanish prospective cohort study.<sup>3</sup> However, a cohort study in the Netherlands reported no association between maternal fish intake during pregnancy and the risk of wheeze in the children from 1 to 8 years of age.<sup>4</sup> In cohort studies in Spain, Germany, and the UK, a protective relationship was found between maternal fish intake during pregnancy and the risk of infantile eczema.<sup>3 5 6</sup> The cohort study in Germany also showed significant

positive associations of maternal intake of margarine and vegetable oils during pregnancy with the risk of eczema during the first two years in children.<sup>5</sup>

Our previous preliminary report found that higher maternal meat intake during pregnancy was significantly associated with an increased risk of suspected atopic eczema, which was based on a physician's diagnosis of atopic eczema or possible atopic eczema, in the infants aged 3-4 months.<sup>7</sup> Using data from the 16- to 24-month follow-up of the cohort with definitions of wheeze and eczema according to the diagnostic criteria of the International Study of Asthma and Allergies in Childhood (ISAAC), we gained further insights into the relation between maternal intake of specific types of fatty acids, cholesterol, meat, and fish during pregnancy and the risk of wheeze and eczema in infants in Japan, where fish intake is high.

## METHODS

### Study population

The Osaka Maternal and Child Health Study (OMCHS) is a prospective cohort study. Details of the OMCHS have been described elsewhere.<sup>8</sup> We recruited pregnant women at baseline. Neyagawa City is one of the 43 municipalities in Osaka Prefecture, a metropolis in Japan. Of the 3639 eligible subjects in Neyagawa City, 627 women (17.2%) participated in the OMCHS between November 2001 and March 2003. In order to increase the sample size, pregnant women living in municipalities other than Neyagawa City were also recruited. Eight pregnant females who did not live in Neyagawa City but who had become aware of the OMCHS at an obstetric clinic before August 2002 decided by themselves to participate in the OMCHS. Also, there were 77 participants who received explanations of the OMCHS from public health nurses in 6 other municipalities from August 2002 to March 2003. From October 2002 to March 2003, 290 participants were recruited from a university hospital and 3 obstetric hospitals in 3 other municipalities.

Finally, a total of 1,002 pregnant women at any stage of pregnancy gave their fully informed consent in writing and completed the baseline survey. Of the 1,002 subjects, 867 mother-child pairs participated in the second survey from 2 to 9 months postpartum. Of the 867 mother-child pairs, 763 participated in the third survey from 16 to 24 months postpartum. The analytic population consisted of these 763 mother-child pairs. The ethics committee of the Osaka City University School of Medicine approved the OMCHS.

### Measurements

At baseline, each participant filled out a set of 2 self-administered questionnaires. Also, a self-administered questionnaire was used in both the second and third survey. Participants mailed these materials to the data management center at the time of each

survey. Research technicians completed missing or illogical data by telephone interview.

In the baseline survey, dietary habits during the preceding month were assessed using a self-administered, semi-quantitative, comprehensive, diet history questionnaire (DHQ). Details of the DHQ's structure, calculation of dietary intake, and validity for commonly studied nutritional factors have been published elsewhere.<sup>9 10</sup> Estimates of daily intake for foods (150 items in total), energy, and selected nutrients were calculated using an ad hoc computer algorithm for the DHQ,<sup>9 10</sup> based on the Standard Tables of Food Composition in Japan.<sup>11 12</sup> Information on dietary supplements was not used in the calculation of dietary intake. The correlation coefficients for nutrient intake between those estimated from the DHQ and those observed by a 3-day dietary record were 0.75, 0.50, 0.37, and 0.49 for saturated fatty acids, monounsaturated fatty acids, polyunsaturated fatty acids, and cholesterol, respectively, in women.<sup>9</sup> A highly positive correlation was also observed between marine-origin n-3 polyunsaturated fatty acid intake estimated by the DHQ and the corresponding concentration in the serum phospholipid fraction in women ( $r = 0.69$ ).<sup>10</sup> Energy-adjusted intake by the residual method was used for the analyses.<sup>13</sup>

A second questionnaire at baseline asked about maternal age, gestation, family income, maternal and paternal education, maternal and paternal history of asthma, atopic eczema, and allergic rhinitis, and changes in diet in the previous one month. A paternal or maternal history of asthma, atopic eczema, and allergic rhinitis was defined as positive if the respective parent had been treated with medications for any of these allergic disorders at some time prior to the start of the survey.

A self-administered questionnaire in the second survey elicited information on the baby's sex, birth weight, and date of birth, number of baby's older siblings, maternal smoking during pregnancy, and smoking in the household. A self-administered questionnaire in the third survey included questions on breastfeeding duration in months and symptoms of wheeze and eczema based on the ISAAC phase-I questionnaire.<sup>14 15</sup> Wheeze was defined as present if the mother answered "yes" to the written question "Has your child had wheezing or whistling in the chest in the last 12 months?". For eczema, affirmative answers to the following three questions were required: "Has your child ever had an itchy rash which was coming and going for at least 6 months?", "Has your child had this itchy rash at any time in the last 12 months?" and "Has this itchy rash at any time affected any of the following places: the folds of the elbow, behind the knees, in front of the ankles, under the buttocks, or around the neck, ears, or eyes?"

### **Statistical analysis**

Intake of dietary factors under investigation was categorized at quartile points based on the distribution in 763 subjects. The following variables were *a priori* selected as

potential confounding factors: maternal age, gestation at baseline, residential municipality, family income, maternal and paternal education, maternal and paternal history of asthma, atopic eczema, and allergic rhinitis, maternal intake of vitamins D and E during pregnancy, changes in maternal diet in the previous 1 month, season when data at baseline were collected, maternal smoking during pregnancy, baby's older siblings, baby's sex, baby's birth weight, household smoking in the same room as the infant, breastfeeding duration, and time of delivery before the third survey.

Logistic regression analysis was used to estimate crude odds ratios (ORs) and 95% confidence intervals (CIs) of wheeze and eczema for each category of dietary intake under study in comparison with the lowest intake category. Multiple logistic regression analysis was employed to control for potential confounding factors. Trend of association was assessed by a logistic regression model assigning consecutive integers (1 to 4) to the quartiles of the exposure variables. All statistical analyses were conducted using the SAS software package version 9.1 (SAS Institute, Inc., Cary, NC).

## RESULTS

Of the 763 infants, the third survey revealed that 169 (22.1%) and 142 (18.6%) had symptoms of wheeze and eczema based on the ISAAC criteria, respectively. About 75% of infants had been breastfed for 6 months or longer (table 1). Slight or substantial changes in diet in the previous 1 month were reported by 551 mothers due to nausea gravidarum (n = 453), maternal and fetal health (n = 89), and other reasons (n = 9). Maternal mean daily total energy intake and energy-adjusted consumption of n-3 and n-6 polyunsaturated fatty acids during pregnancy were 7629 kJ, 2.4 g, and 11.5 g, respectively (table 2).

**Table 1** Distribution of selected characteristics of 763 parent-child pairs

Variable	n (%) or mean (SD)
Baseline characteristics	
Maternal age (years)	30.0 (4.0)
Gestation (weeks)	17.7 (6.7)
Residential municipality	
Neyagawa City	457 (59.9)
Other than Neyagawa City	306 (40.1)
Family income (% yen/year)	
< 4,000,000	211 (27.7)
4,000,000–5,999,999	313 (41.0)
6,000,000+	239 (31.3)
Maternal education (% years)	
< 13	212 (27.8)

13–14	329 (43.1)
15+	222 (29.1)
Paternal education (% years)	
< 13	289 (37.9)
13–14	127 (16.6)
15+	347 (45.5)
Maternal history of asthma (%)	80 (10.5)
Maternal history of atopic eczema (%)	120 (15.7)
Maternal history of allergic rhinitis (%)	256 (33.6)
Paternal history of asthma (%)	64 (8.4)
Paternal history of atopic eczema (%)	72 (9.4)
Paternal history of allergic rhinitis (%)	143 (18.7)
Changes in diet in the previous 1 month (%)	
None or seldom	212 (27.8)
Slight	336 (44.0)
Substantial	215 (28.2)
Season when data were collected (%)	
Spring	241 (31.6)
Summer	127 (16.6)
Fall	167 (21.9)
Winter	228 (29.9)
Characteristics at postnatal assessment	
Maternal smoking during pregnancy (%)	97 (12.7)
Baby's older siblings (% 1 or more)	381 (49.9)
Baby's sex (% male)	403 (52.8)
Baby's birth weight (g)	3071.0 (415.5)
Household smoking in same room as infant	195 (25.6)
Breastfeeding duration (% months)	
< 6	189 (24.8)
6+	574 (75.2)
Time of delivery before third survey (% months)	
16–19	263 (34.5)
20	370 (48.5)
21–24	130 (17.0)

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**Table 2** Distribution of daily nutrients and food intake in 763 pregnant women at baseline\*

Variable	Mean (SD)
Daily nutrient intake	
Total energy (kJ)	7628.6 (1924.1)
Total fat (g)	61.6 (10.8)
Saturated fatty acids (g)	17.4 (4.0)
Monounsaturated fatty acids (g)	21.3 (4.9)
n-3 Polyunsaturated fatty acids (g)	2.4 (0.7)
$\alpha$ -Linolenic acid (g)	1.8 (0.6)
Eicosapentaenoic acid (g)	0.17 (0.11)
Docosahexaenoic acid (g)	0.30 (0.17)
n-6 Polyunsaturated fatty acids (g)	11.5 (2.7)
Linoleic acid (g)	11.3 (2.6)
Arachidonic acid (g)	0.14 (0.04)
Cholesterol (mg)	308.8 (111.4)
Vitamin E (mg)	7.8 (1.7)
Vitamin D ( $\mu$ g)	6.2 (3.7)
Daily intake	
Meat (g)	60.2 (28.8)
Fish (g)	48.4 (26.5)

\*Nutrient and food intake were adjusted for total energy intake using the residual method.

Table 3 presents ORs and 95% CIs for the risk of wheeze and eczema in the offspring according to maternal dietary intake of specific types of fatty acids and cholesterol during pregnancy. After adjustment for confounding factors under study, maternal intake of total fat, saturated fatty acids, monounsaturated fatty acids, and n-3 polyunsaturated fatty acids during pregnancy was not measurably related to the risk of wheeze or eczema in the offspring. Compared with maternal  $\alpha$ -linolenic acid intake during pregnancy in the first quartile, such intake in the fourth quartile was independently associated with a reduced risk of wheeze in the offspring (adjusted OR = 0.52 [95% CI: 0.28–0.97]) although the inverse exposure-response relationship was not statistically significant. There was no association between maternal  $\alpha$ -linolenic acid intake during pregnancy and the risk of infantile eczema. Maternal eicosapentaenoic

acid intake during pregnancy was not related to the risk of childhood wheeze or eczema. Maternal consumption of docosahexaenoic acid in the second and fourth quartiles was independently related to a decreased risk of wheeze in the infants although the inverse linear trend was not statistically significant (adjusted OR between extreme quartiles = 0.37 [95% CI: 0.15–0.91]). No such relationship was detected for infantile eczema. Maternal intake of n-6 polyunsaturated fatty acids and linoleic acid during pregnancy in the highest quartile was independently associated with an increased risk of infantile eczema, but not wheeze, showing clear inverse exposure-response relationships (adjusted ORs = 2.25 and 2.11 [95% CIs: 1.13–4.54 and 1.06–4.26], p for trend = 0.01 and 0.03, respectively). No statistically significant exposure-response relationships were observed between maternal intake of arachidonic acid and cholesterol and the ratio of n-3 to n-6 polyunsaturated fatty acid consumption during pregnancy and the risk of wheeze or eczema in the offspring after multivariate adjustment.

After adjustment for potential confounders excluding maternal intake of vitamins D and E during pregnancy, inverse exposure-response associations between maternal intake of total fat, saturated fatty acids, monounsaturated fatty acids, n-3 polyunsaturated fatty acids, and  $\alpha$ -linolenic acid during pregnancy and the risk of wheeze in infants were statistically significant whereas maternal intake of n-6 polyunsaturated fatty acids and linoleic acid during pregnancy was not related to the risk of infantile eczema.

There were no evident relationships between maternal intake of meat and fish during pregnancy and the risk of wheeze or fish in the offspring (table 4).

**Table 3** Odds ratios and 95% confidence intervals of wheeze and eczema in 763 infants aged 16-24 months by quartiles of maternal consumption of specific fats during pregnancy

Variable*	Wheeze			Eczema		
	No. cases	Crude OR (95% CI)	Adjusted OR (95% CI) <sup>†</sup>	No. cases	Crude OR (95% CI)	Adjusted OR (95% CI) <sup>†</sup>
<b>Total fat</b>						
Q1 (50.3)	49	1.00	1.00	34	1.00	1.00
Q2 (58.5)	52	1.08 (0.68–1.70)	1.12 (0.68–1.85)	39	1.18 (0.71–1.97)	1.55 (0.88–2.76)
Q3 (64.2)	30	0.54 (0.32–0.89)	0.59 (0.33–1.04)	33	0.96 (0.56–1.63)	1.37 (0.75–2.52)
Q4 (72.4)	38	0.72 (0.44–1.16)	0.80 (0.44–1.43)	36	1.07 (0.63–1.80)	1.49 (0.79–2.83)
p for trend		0.03	0.18		0.98	0.30
<b>Saturated fatty acids</b>						
Q1 (13.3)	55	1.00	1.00	33	1.00	1.00
Q2 (15.9)	41	0.67 (0.42–1.07)	0.84 (0.51–1.39)	34	1.03 (0.61–1.75)	1.38 (0.78–2.48)
Q3 (18.3)	37	0.59 (0.36–0.95)	0.66 (0.39–1.10)	34	1.03 (0.61–1.75)	1.32 (0.74–2.37)
Q4 (21.7)	36	0.57 (0.35–0.92)	0.65 (0.39–1.10)	41	1.30 (0.78–2.18)	1.60 (0.92–2.83)
p for trend		0.02	0.07		0.33	0.13
<b>Monounsaturated fatty acids</b>						
Q1 (16.7)	47	1.00	1.00	40	1.00	1.00
Q2 (19.9)	50	1.08 (0.68–1.71)	1.21 (0.73–2.02)	32	0.76 (0.45–1.26)	0.90 (0.51–1.59)
Q3 (22.2)	36	0.71 (0.43–1.15)	0.92 (0.52–1.61)	31	0.73 (0.43–1.22)	1.03 (0.56–1.88)
Q4 (25.7)	36	0.71 (0.43–1.15)	0.79 (0.43–1.45)	39	0.96 (0.59–1.58)	1.32 (0.70–2.49)
p for trend		0.06	0.33		0.85	0.36
<b>n-3 Polyunsaturated fatty acids</b>						
Q1 (1.7)	53	1.00	1.00	35	1.00	1.00
Q2 (2.2)	38	0.64 (0.40–1.03)	0.57 (0.33–0.99)	38	1.10 (0.66–1.84)	1.72 (0.95–3.13)

Q3 (2.5)	42	0.73 (0.46–1.16)	0.66 (0.36–1.22)	33	0.93 (0.55–1.56)	1.63 (0.83–3.22)
Q4 (3.0)	36	0.60 (0.37–0.97)	0.53 (0.26–1.08)	36	1.03 (0.61–1.73)	1.74 (0.82–3.73)
p for trend		0.06	0.13		0.92	0.20
$\alpha$ -Linolenic acid						
Q1 (1.3)	53	1.00	1.00	35	1.00	1.00
Q2 (1.6)	38	0.64 (0.40–1.03)	0.63 (0.37–1.07)	33	0.93 (0.55–1.56)	1.28 (0.71–2.29)
Q3 (1.9)	45	0.80 (0.50–1.26)	0.78 (0.45–1.35)	35	0.99 (0.59–1.67)	1.69 (0.91–3.13)
Q4 (2.3)	33	0.54 (0.33–0.88)	0.52 (0.28–0.97)	39	1.14 (0.68–1.90)	1.79 (0.93–3.50)
p for trend		0.04	0.08		0.58	0.06
Eicosapentaenoic acid						
Q1 (0.08)	48	1.00	1.00	39	1.00	1.00
Q2 (0.13)	39	0.76 (0.47–1.23)	0.77 (0.42–1.41)	36	0.90 (0.54–1.49)	0.99 (0.51–1.89)
Q3 (0.18)	38	0.74 (0.45–1.19)	0.76 (0.37–1.58)	33	0.81 (0.48–1.35)	1.12 (0.52–2.45)
Q4 (0.28)	44	0.89 (0.55–1.42)	0.76 (0.33–1.80)	34	0.84 (0.50–1.40)	0.98 (0.39–2.50)
p for trend		0.59	0.58		0.44	0.95
Docosahexaenoic acid						
Q1 (0.15)	55	1.00	1.00	40	1.00	1.00
Q2 (0.24)	29	0.44 (0.26–0.72)	0.41 (0.20–0.81)	40	0.99 (0.61–1.63)	1.50 (0.76–3.02)
Q3 (0.32)	50	0.87 (0.55–1.37)	0.72 (0.33–1.57)	32	0.76 (0.45–1.26)	1.11 (0.49–2.54)
Q4 (0.46)	35	0.55 (0.34–0.89)	0.37 (0.15–0.91)	30	0.70 (0.41–1.18)	0.86 (0.33–2.28)
p for trend		0.12	0.14		0.11	0.57
n-6 Polyunsaturated fatty acids						
Q1 (9.0)	48	1.00	1.00	34	1.00	1.00
Q2 (10.7)	39	0.76 (0.47–1.23)	0.83 (0.49–1.42)	30	0.86 (0.50–1.46)	1.18 (0.65–2.17)
Q3 (12.1)	44	0.89 (0.55–1.42)	0.93 (0.53–1.64)	37	1.10 (0.66–1.85)	1.81 (0.97–3.42)
Q4 (14.1)	38	0.74 (0.45–1.19)	0.80 (0.42–1.54)	41	1.25 (0.76–2.09)	2.25 (1.13–4.54)

p for trend		0.32		0.61		0.25		0.01
Linoleic acid								
Q1 (8.8)	48	1.00		1.00	34	1.00		1.00
Q2 (10.4)	40	0.78 (0.48–1.26)		0.85 (0.50–1.45)	32	0.92 (0.54–1.57)		1.22 (0.67–2.21)
Q3 (11.8)	43	0.86 (0.54–1.38)		0.87 (0.49–1.53)	35	1.03 (0.61–1.74)		1.58 (0.85–2.98)
Q4 (13.8)	38	0.74 (0.45–1.19)		0.79 (0.41–1.50)	41	1.25 (0.76–2.09)		2.11 (1.06–4.26)
p for trend		0.28		0.51		0.33		0.03
Arachidonic acid								
Q1 (0.09)	50	1.00		1.00	49	1.00		1.00
Q2 (0.12)	45	0.86 (0.54–1.37)		0.98 (0.58–1.67)	29	0.52 (0.31–0.85)		0.55 (0.31–0.98)
Q3 (0.15)	37	0.67 (0.41–1.09)		0.74 (0.42–1.30)	34	0.62 (0.38–1.02)		0.73 (0.41–1.30)
Q4 (0.19)	37	0.67 (0.41–1.09)		0.77 (0.43–1.37)	30	0.54 (0.32–0.89)		0.69 (0.38–1.27)
p for trend		0.06		0.26		0.03		0.40
n-3/n-6 Polyunsaturated fatty acid ratio								
Q1 (0.17)	46	1.00		1.00	31	1.00		1.00
Q2 (0.19)	44	0.94 (0.58–1.50)		0.88 (0.52–1.51)	33	1.07 (0.63–1.84)		1.24 (0.68–2.28)
Q3 (0.21)	37	0.75 (0.46–1.22)		0.69 (0.39–1.23)	46	1.63 (0.98–2.72)		2.13 (1.17–3.96)
Q4 (0.24)	42	0.88 (0.55–1.42)		0.81 (0.42–1.55)	32	1.03 (0.60–1.78)		1.32 (0.65–2.71)
p for trend		0.44		0.38		0.52		0.18
Cholesterol								
Q1 (200.2)	53	1.00		1.00	51	1.00		1.00
Q2 (263.0)	34	0.56 (0.34–0.91)		0.65 (0.38–1.10)	30	0.51 (0.30–0.84)		0.53 (0.30–0.92)
Q3 (333.5)	41	0.71 (0.44–1.13)		0.81 (0.48–1.37)	29	0.49 (0.29–0.81)		0.56 (0.31–0.99)
Q4 (430.7)	41	0.71 (0.44–1.13)		0.85 (0.49–1.47)	32	0.55 (0.33–0.90)		0.71 (0.39–1.27)
p for trend		0.25		0.74		0.02		0.27

\*Quartile medians in g/day (except for cholesterol, mg/day) adjusted energy intake using the residual method are given in parentheses,

except for the ratio of n-3 to n-6 polyunsaturated fatty acids, which was based on crude intake in g/day.

†Adjustment for maternal age, gestation at baseline, residential municipality at baseline, family income, maternal and paternal education, maternal and paternal history of asthma, atopic eczema, and allergic rhinitis, maternal intake of vitamins D and E during pregnancy, changes in maternal diet in the previous 1 month, season when data at baseline were collected, maternal smoking during pregnancy, baby's older siblings, baby's sex, baby's birth weight, household smoking in same room as infant, breastfeeding duration, and time of delivery before third survey.

**Table 4** Odds ratios and 95% confidence intervals of wheeze and eczema in 763 infants aged 16-24 months by quartiles of maternal consumption of meat and fish during pregnancy

Variable*	Wheeze			Eczema		
	No. cases	Crude OR (95% CI)	Adjusted OR (95% CI) <sup>†</sup>	No. cases	Crude OR (95% CI)	Adjusted OR (95% CI) <sup>†</sup>
<b>Meat</b>						
Q1 (33.8)	54	1.00	1.00	40	1.00	1.00
Q2 (49.0)	37	0.61 (0.37–0.97)	0.67 (0.40–1.11)	26	0.59 (0.34–1.01)	0.68 (0.38–1.21)
Q3 (63.6)	35	0.57 (0.35–0.91)	0.57 (0.33–0.95)	32	0.76 (0.45–1.26)	0.80 (0.46–1.39)
Q4 (90.8)	43	0.73 (0.46–1.16)	0.77 (0.47–1.27)	44	1.12 (0.69–1.83)	1.31 (0.78–2.22)
p for trend		0.16	0.22		0.47	0.28
<b>Fish</b>						
Q1 (23.4)	49	1.00	1.00	39	1.00	1.00
Q2 (38.7)	41	0.79 (0.49–1.26)	0.88 (0.49–1.58)	35	0.87 (0.52–1.44)	1.10 (0.59–2.05)
Q3 (51.7)	39	0.74 (0.46–1.19)	0.68 (0.34–1.36)	40	1.03 (0.62–1.69)	1.33 (0.65–2.72)
Q4 (73.2)	40	0.76 (0.47–1.23)	0.67 (0.30–1.48)	28	0.67 (0.39–1.13)	0.73 (0.30–1.75)
p for trend		0.25	0.28		0.24	0.68

\*Quartile medians in g/day adjusted energy intake using the residual method are given in parentheses.

<sup>†</sup>Adjustment for maternal age, gestation at baseline, residential municipality at baseline, family income, maternal and paternal education, maternal and paternal history of asthma, atopic eczema, and allergic rhinitis, maternal intake of vitamins D and E during pregnancy, changes in maternal diet in the previous 1 month, season when data at baseline were collected, maternal smoking during pregnancy, baby's older siblings, baby's sex, baby's birth weight, household smoking in same room as infant, breastfeeding duration, and time of delivery before third survey.

## DISCUSSION

During immune maturation, naive Th0 cells are able to modulate the predominant expression of a Th1- or Th2-like cytokine pattern according to numerous eicosanoids and cytokines synthesized by antigen-presenting cells.<sup>16</sup> Polyunsaturated fatty acids and their derivatives can influence the Th1:Th2 ratio. An intervention study among atopic pregnant Australian women demonstrated that fish oil supplementation achieved a significant increase in n-3 polyunsaturated fatty acids in neonatal erythrocyte membranes compared with the control group and that the neonatal in vitro IL-10 response to cat allergen was significantly lower in the fish oil-supplemented group than in the control group.<sup>17</sup> Other reports of that intervention study showed that IL-13 levels in cord plasma and neonatal leukotriene B<sub>4</sub> production by cord blood-derived neutrophils were significantly lower in infants from the intervention group than from the placebo group.<sup>18 19</sup> In a randomized clinical trial of 528 pregnant Danish women, the risk of allergic asthma, atopic dermatitis, or allergic rhinitis during the first 15-16 years of life was significantly lower in children whose mothers had received supplements of fish oil (2.7 g marine n-3 polyunsaturated fatty acids per day) compared to those in the placebo group (olive oil) although the risk in children of mothers in the no oil group was remarkably similar to the risk in children whose mothers had received fish oil.<sup>20</sup> These findings agree with our results in relation to maternal intake of  $\alpha$ -linolenic acid and docosahexaenoic acid during pregnancy and the risk of infantile wheeze. The lack of association of wheeze in offspring with maternal eicosapentaenoic acid intake in the present study might to some extent be ascribed to the fact that docosahexaenoic acid is preferentially transferred to the fetus from the maternal circulation.<sup>21</sup> A UK prospective study showed that the levels of 11 individual n-3 and n-6 fatty acids in both cord blood red cells and maternal blood red cells obtained after 20 weeks of pregnancy were not related to the risk of infantile wheezing and eczema.<sup>22</sup> Japanese people consume a large quantity of fish compared to people in Western countries, but the median value of daily marine origin n-3 polyunsaturated fatty acid intake in the present study was 0.48 g, which was much lower than the supplemented quantity in the above-cited Danish intervention study.<sup>20</sup> A clear protective relationship between maternal intake of marine origin n-3 polyunsaturated fatty acids and the risk of infantile atopic eczema might be substantiated if consumption were much higher than in this study. The nonsignificant association of wheeze and eczema in the offspring with maternal fish intake in the current study could be due to unrecognized active agents in fish that interfered with the expected benefit to offspring of maternal fish intake during pregnancy. For example, methylmercury and dioxins are accumulated in fish and shellfish through the marine food web. This hypothetical adverse effect might not be detected in Western populations where fish intake is low.<sup>23 5 6</sup>

Linoleic acid is converted into arachidonic acid, which enhances inflammatory processes via its derivatives prostaglandin E<sub>2</sub> and leukotriene B<sub>4</sub>. Our results regarding maternal intake of n-6 polyunsaturated fatty acids and linoleic acid and infantile eczema might be reasonable. On the other hand, lipoxins, derived from arachidonic acid, decrease the synthesis of TNF- $\alpha$  and leucocyte chemotaxis.<sup>16</sup> Arachidonic acid as well as docosahexaenoic acid is essential to the placenta and the fetus.<sup>16</sup> A US birth cohort study found that both higher levels of cord blood eicosapentaenoic acid and arachidonic acid were associated with attenuation of cord blood lymphocyte proliferation and decreased IFN- $\gamma$  secretion in response to allergen stimulation whereas higher levels of linoleic acid were correlated with increased allergen-stimulated IL-13 levels.<sup>23</sup> Given that the beneficial effect of arachidonic acid on fetal immune development counteracts the detrimental effect, the present findings on null associations between maternal intake of n-6 polyunsaturated fatty acids, linoleic acid, and arachidonic acid during pregnancy and infantile wheeze also might be reasonable.

The present study had methodological strengths. Study subjects were homogeneous in terms of having the same residential background. Extensive data on potential confounding factors were controlled for. Especially, maternal intake of vitamins D and E during pregnancy was the most important confounders. Such intake was inversely related to the risk of infantile wheeze and eczema. The prospective design was likely to reduce the possibility of recall bias. The definition of wheeze and eczema was based on the ISAAC questions. However, the outcomes under investigation were measured at varying ages between 16 and 24 months and additionally wheeze in infancy is known not to be a good predictor of asthma over the longer term. The consequence would have been an underestimation of values in our results.

Other weaknesses of the current study should be borne in mind. Our DHQ could only approximate consumption. The consequence would bias the estimates of the observed association toward the null. Our DHQ was designed to assess recent dietary intake, i.e. for 1 month prior to completing the questionnaire. This disadvantage is likely to be alleviated after adjustment for the season when data were collected, however. Changes in diet in the past 1 month were controlled for because pregnant females are likely to change their diet.

The follow-up rate was not good. Of the 1002 participants at baseline, 239 mother-child pairs did not take part in the third survey. There were no material differences between the 239 non-participants and the 763 participants in the third survey with regard to distribution of maternal age, maternal and paternal history of asthma, atopic eczema, and allergic rhinitis, and maternal intake of total energy, monounsaturated fatty acids, n-3 polyunsaturated fatty acids, n-6 polyunsaturated fatty acids other than arachidonic acid, meat, and fish. Compared with non-participants in the third survey, participants were less likely to report low family income and a low maternal and paternal educational level and were more likely to have a high intake of

total fat, saturated fatty acids, cholesterol, arachidonic acid, and vitamins D and E. Moreover, at baseline, the participation rate in Neyagawa City was only 17.2% and in other areas the participation rate could not be calculated because the exact number of eligible subjects among the sources from which participants were recruited was not available. Thus, the mother-child pairs in this study were likely not representative of Japanese mother-child pairs in the general population. In fact, educational levels were higher in the mothers in our study than in the general population. According to the 2000 population census of Japan, the proportions of females aged 30 to 34 years in Osaka Prefecture with years of education of < 13, 13–14, 15+, and unknown were 49.2, 32.3, 13.6, and 4.9%, respectively.<sup>24</sup> The present population might have had a greater awareness about health than the general population.

The current finding regarding the null association between maternal meat intake during pregnancy and infantile eczema was at variance with our previous results showing a positive association between maternal meat intake during pregnancy and suspected atopic eczema in the infants aged 3–4 months. The definition of suspected atopic eczema might have been crude. Of 58 infants identified with suspected atopic eczema, 25 infants (43.1%) were classified as having eczema based on the ISAAC criteria in the third survey. In view of the multiple analyses and the *p* values, it is possible that some, or all, of the findings in the current study and in our previous study occurred by chance.

Our results suggest that maternal intake of  $\alpha$ -linolenic acid and docosahexaenoic acid during pregnancy may be preventive against infantile wheeze whereas maternal intake of n-6 polyunsaturated fatty acids, especially linoleic acid, during pregnancy may increase the risk of childhood eczema. Further epidemiological investigations are required to ascertain whether the relationships observed in the current study are replicated in other populations.

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## REFERENCES

1. Devereux G, Seaton A. Diet as a risk factor for atopy and asthma. *J Allergy Clin Immunol* 2005; 115: 1109-17.
2. Salam MT, Li YF, Langholz B, et al. Maternal fish consumption during pregnancy and risk of early childhood asthma. *J Asthma* 2005; 42: 513-8.
3. Romieu I, Torrent M, Garcia-Esteban R, et al. Maternal fish intake during pregnancy and atopy and asthma in infancy. *Clin Exp Allergy* 2007; 37: 518-25.
4. Willers SM, Wijga AH, Brunekreef B, et al. Maternal food consumption during pregnancy and the longitudinal development of childhood asthma. *Am J Respir Crit Care Med* 2008; 178: 124-31.
5. Sausenthaler S, Koletzko S, Schaaf B, et al. Maternal diet during pregnancy in relation to eczema and allergic sensitization in the offspring at 2 y of age. *Am J Clin Nutr* 2007; 85: 530-7.
6. Willers SM, Devereux G, Craig LCA, et al. Maternal food consumption during pregnancy and asthma, respiratory and atopic symptoms in 5-year-old children. *Thorax* 2007; 62: 773-9.
7. Saito K, Yokoyama T, Miyake Y, et al. Maternal meat and fat consumption during pregnancy and suspected atopic eczema in Japanese infants aged 3-4 months: The Osaka Maternal and Child Health Study. *Pediatr Allergy Immunol* in press.
8. Miyake Y, Tanaka K, Sasaki S, et al. Breastfeeding and atopic eczema in Japanese infants: The Osaka Maternal and Child Health Study. *Pediatr Allergy Immunol* 2009; 20: 234-41.
9. Sasaki S, Yanagibori R, Amano K. Self-administered diet history questionnaire developed for health education: a relative validation of the test-version by comparison with 3-day diet record in women. *J Epidemiol* 1998; 8: 203-15.
10. Sasaki S, Ushio F, Amano K, et al. Serum biomarker-based validation of a self-administered diet history questionnaire for Japanese subjects. *J Nutr Sci Vitaminol* 2000; 46: 285-96.
11. Science and Technology Agency. Standard Tables of Food Composition in Japan. 5th revised and enlarged ed. Tokyo: Printing Bureau of the Ministry of Finance, 2005 (in Japanese).
12. Science and Technology Agency. Standard Tables of Food Composition in Japan, fatty acids section. 5th revised and enlarged ed. Tokyo: Printing Bureau of the Ministry of Finance, 2005 (in Japanese).
13. Willett W, Stampfer MJ. Total energy intake: implications for epidemiologic analyses. *Am J Epidemiol* 1986; 124: 17-27.
14. The International Study of Asthma and Allergies in Childhood (ISAAC) Steering Committee. Worldwide variations in the prevalence of asthma symptoms: the International Study of Asthma and Allergies in Childhood (ISAAC). *Eur Respir J* 1998; 12: 315-35.

15. Williams H, Robertson C, Stewart A, et al. Worldwide variations in the prevalence of symptoms of atopic eczema in the international study of asthma and allergies in childhood. *J Allergy Clin Immunol* 1999; 103: 125-38.
16. Enke U, Seyfarth L, Schleussner E, et al. Impact of PUFA on early immune and fetal development. *Br J Nutr* 2008; 100: 1158-68.
17. Dunstan JA, Mori TA, Barden A, et al. Fish oil supplementation in pregnancy modifies neonatal allergen-specific immune responses and clinical outcomes in infants at high risk of atopy: a randomized, controlled trial. *J Allergy Clin Immunol* 2003; 112: 1178-84.
18. Dunstan JA, Mori TA, Barden A, et al. Maternal fish oil supplementation in pregnancy reduces interleukin-13 levels in cord blood of infants at high risk of atopy. *Clin Exp Allergy* 2003; 33: 442-8.
19. Prescott SL, Barden AE, Mori TA, et al. Maternal fish oil supplementation in pregnancy modifies neonatal leukotriene production by cord-blood-derived neutrophils. *Clin Sci (Lond)* 2007; 113: 409-16.
20. Olsen SF, Østerdal ML, Salvig JD, et al. Fish oil intake compared with olive oil intake in late pregnancy and asthma in the offspring: 16 y of registry-based follow-up from a randomized controlled trial. *Am J Clin Nutr* 2008; 88: 167-75.
21. Blümer N, Renz H. Consumption of omega3-fatty acids during perinatal life: role in immuno-modulation and allergy prevention. *J Perinat Med* 2007; 35 (suppl): S12-8.
22. Newson RB, Shaheen SO, Henderson AJ, et al. Umbilical cord and maternal blood red cell fatty acids and early childhood wheezing and eczema. *J Allergy Clin Immunol* 2004; 114: 531-7.
23. Gold DR, Willwerth BM, Tantisira KG, et al. Associations of cord blood fatty acids with lymphocyte proliferation, IL-13, and IFN- $\gamma$ . *J Allergy Clin Immunol* 2006; 117: 931-8.
24. Statistic Bureau, Ministry of Public Management, Home Affairs, Posts and Telecommunications. 2000 population census of Japan, Vol. 3-2-27, labour force status of population, industry (major groups) of employed persons, and education. Osaka-fu. Tokyo: Statistic Bureau, Ministry of Public Management, Home Affairs, Posts and Telecommunications, 2002 (in Japanese).