

ORIGINAL ARTICLE

Intake of antioxidants during pregnancy and the risk of allergies and asthma in the offspring

BI Nwaru¹, M Erkkola², S Ahonen^{1,3}, M Kaila^{4,5}, C Kronberg-Kippilä⁶, J Ilonen^{7,8}, O Simell⁹,
M Knip^{4,5,10,11}, R Veijola¹² and SM Virtanen^{1,3,4,5,6}

¹School of Health Sciences, University of Tampere, Tampere, Finland; ²Division of Nutrition, Department of Food and Environmental Sciences, University of Helsinki, Helsinki, Finland; ³The Science Center of Pirkanmaa Hospital District, Tampere, Finland; ⁴Department of Pediatrics, Tampere University Hospital, Tampere, Finland; ⁵Hjelt Institute, University of Helsinki, Helsinki, Finland; ⁶Nutrition Unit, Department of Lifestyle and Participation, National Institute for Health and Welfare, Helsinki, Finland; ⁷Immunogenetics Laboratory, University of Turku, Turku, Finland; ⁸Department of Clinical Microbiology, University of Eastern Finland, Kuopio, Finland; ⁹Department of Pediatrics, University of Turku, Turku, Finland; ¹⁰Hospital for Children and Adolescents, University of Helsinki and Helsinki University Central Hospital, Helsinki, Finland; ¹¹Folkhälsan Research Center, Helsinki, Finland and ¹²Department of Pediatrics, University of Oulu, Oulu, Finland

Background/Objectives: The potential immune functions related to the damages induced by oxygen-free radicals suggest that antioxidants may have a role in the development of allergies. The objective was to investigate the association between maternal intake of antioxidants during pregnancy and the risk of asthma, rhinitis and eczema in 5-year-old children.

Subjects/Methods: This study was on the basis of the Finnish Type 1 Diabetes Prediction and Prevention Nutrition Study, a population-based birth cohort study with 5-year follow-up. Complete information on maternal food frequency questionnaire data and ISAAC (International Study of Asthma and Allergies in Childhood)-based allergic outcomes were available for 2441 children. Cox proportional regression and logistic regression were used for the analyses.

Results: Maternal intake of any of the antioxidants was not significantly associated with the risk of asthma, rhinitis or eczema in the offspring, except for dietary intake of magnesium, which was independently associated with protection against eczema (OR 0.78; 95% CI 0.62–0.97).

Conclusion: Maternal intake of dietary magnesium during pregnancy may protect against the risk of eczema in the offspring. We did not confirm previous observations concerning other antioxidants. This may be due to the variable amount of antioxidant intake across studies and also indicative of the hypothesis that there may be a critical time window in pregnancy during which antioxidants might modify the risk of allergies in the offspring.

European Journal of Clinical Nutrition advance online publication, 11 May 2011; doi:10.1038/ejcn.2011.67

Keywords: allergies; antioxidants; asthma; children; maternal diet; pregnancy

Correspondence: BI Nwaru, Tampere School of Public Health, University of Tampere, Medisiinäinkatu 3, Tampere FIN-33014, Finland.
E-mail: bright.nwaru@uta.fi

Contributors: BIN and ME designed, analyzed and prepared the present manuscript. SMV designed and is responsible for the DIPP Nutrition and Allergy studies. SMV and MK have planned the allergy study questionnaire for 5-year olds. SA and CK-K were responsible for data acquisition and preparation, interpretation of results, and critically reviewing the manuscript. JI, RV, OS, and MK were responsible for data collection in the clinics and for pediatric expertise in this study, and review of the manuscript and revising it critically for important intellectual content. All authors have commented and approved the final version.

Received 22 November 2010; revised 25 February 2011; accepted 1 April 2011

Introduction

Allergic disorders are a consequence of immune processes that promote a shift in the balance between the Th1 and Th2 cytokines, favoring Th2 dominance (Kay, 2001). This process is believed to result in the production of oxygen-free radicals (Seaton *et al.*, 1994). In addition to its natural intra- and extracellular antioxidant defense systems, the lung depends on nutritional antioxidants to inhibit the propagation of the damages from these free radicals (Seaton *et al.*, 1994; Li-Weber *et al.*, 2002). *In vitro* studies suggest that the deficiency in dietary antioxidants may result in T-helper cell

differentiation and the promotion of the Th2 cell types (Li-Weber *et al.*, 2002). Consequently, it has been hypothesized that inadequate intake of dietary antioxidants may contribute to the increasing frequency of allergies and asthma in industrialized countries (Seaton *et al.*, 1994).

Childhood risk of allergies may already be influenced *in utero* by the antioxidants status of the mother during pregnancy (Prescott *et al.*, 1999; Devereux, 2007). Some studies have reported a reduced risk of asthma and wheezing in the offspring with high maternal intake of vitamin E during pregnancy (Martindale *et al.*, 2005; Devereux *et al.*, 2006; Litonjua *et al.*, 2006; Miyake *et al.*, 2010a), whereas no association was observed regarding eczema (Devereux *et al.*, 2006; Litonjua *et al.*, 2006; Miyake *et al.*, 2010b). Similar inconsistencies have been seen concerning maternal intake of zinc and the risk of allergic outcomes (Shahen *et al.*, 2004; Martindale *et al.*, 2005; Devereux *et al.*, 2006; Litonjua *et al.*, 2006; Miyake *et al.*, 2010b). Umbilical cord selenium and iron has been associated with wheezing and eczema in children (Shahen *et al.*, 2004). Maternal supplementation of folic acid during pregnancy was reported recently to be associated with increased risk of asthma (Whitrow *et al.*, 2009) and wheezing (Håberg *et al.*, 2009), whereas high intake of calcium and β -carotene reduced the risk of allergies in the offspring (Miyake *et al.*, 2010a,b). No material associations have been reported with other antioxidants.

Despite the plausibility of the hypothesis linking maternal intake of antioxidants during pregnancy to the risk of allergies in the offspring, the limited and inconsistent findings reported indicate the need for more accumulated evidence in this area to decipher the role of antioxidants in the occurrence of allergic disorders. The aim of the present study was to prospectively investigate whether maternal intake of antioxidants during pregnancy is associated with the incidence of asthma, rhinitis and the prevalence of eczema by the age of 5 years.

Subjects and methods

Subjects

The Finnish Type 1 Diabetes Prediction and Prevention study, which started in 1994, is a multidisciplinary population-based prospective cohort study that examines potential means to predict and prevent the manifestation of type 1 diabetes (Kupila *et al.*, 2001). Infants born with human leucocyte antigen conferred susceptibility to type 1 diabetes are recruited from three university hospitals in Finland (Turku, Oulu and Tampere) and monitored at 3- to 12-month intervals for diabetes-associated antibodies, growth and environmental exposures. The study procedures were approved by the local ethics committees.

In September 1996 and October 1997, the Finnish Type 1 Diabetes Prediction and Prevention Nutrition Study was started within the framework of the Finnish Type 1 Diabetes Prediction and Prevention study in Oulu (Northern Finland)

and Tampere (Southern Finland), respectively (Virtanen *et al.*, 2006). That study examines the relation of maternal diet during pregnancy and lactation, and the child's diet during infancy to the development of type 1 diabetes, allergies and asthma in childhood. At the age of 5 years, 3523 children who were still participating in the dietary follow-up (born between 20 October 1997 and 29 February 2004) were invited to take part in the allergy study by completing a questionnaire on the child's history of allergic symptoms. Altogether, 3253 (92% of those invited) participated in the study. For the present analysis, complete information on maternal diet during pregnancy and history of allergic outcomes was available for 2441 children (69% of those invited to the allergy study and 75% of those who participated).

Dietary assessment

Maternal food consumption during the eighth month of pregnancy was assessed by means of a validated 181-item food frequency questionnaire (FFQ) (Erkkola *et al.*, 2001) designed specially to reflect the diet of Finnish women of child-bearing age. The FFQ was validated against 10-day food records in a sample of pregnant Finnish women (Erkkola *et al.*, 2001). The validity of the FFQ for antioxidant nutrients was, in general, good: energy adjusted and attenuated correlation coefficients between the two methods ranged from 0.32 for copper to 0.71 for retinol. The only exception was vitamin E for which the correlation was only 0.22.

Mothers received the FFQ by mail after delivery and recorded the foods they ate during the month preceding their pregnancy leave, that is, the eighth month of pregnancy. The FFQ forms were returned to the study center at the child's 3-month visit and were checked by a trained nutritionist. If there were ≥ 10 food items with missing frequency, the FFQ was rejected. Software of the National Institute for Health and Welfare (Helsinki, Finland) was used to convert the food consumption data to each mother's daily nutrient intake, taking into account women's individual choices of fats used in food preparations and baking, and oils used for salad dressing (Arkkola *et al.*, 2006).

Information was also requested concerning the use of supplements during the whole pregnancy period, including the type, brand, and manufacturer of all the supplements used, the amount of each supplement used per day or week, and the weeks of pregnancy during which each supplement was used. The nutrient contents of the supplements registered as drugs were obtained from the Finnish pharmacopoeia. Information on other supplements was acquired from the National Food Administration, the manufacturer or both. Average daily intakes of supplements were calculated as separate variables by in-house software of the National Institute of Health and Welfare.

Assessment of allergic outcomes

At the age of 5 years, families of the participating children completed a questionnaire modified from the International

Study of Asthma and Allergies in Childhood (ISAAC) questionnaire on the child's history of allergic symptoms and asthma (Asher *et al.*, 1995; Remes *et al.*, 1998). Asthma was defined as doctor-diagnosed asthma plus either any wheezing symptom during the preceding 12 months or use of asthma medication during the preceding 12 months. Age of the child at asthma diagnosis was determined by the question: 'at what age was asthma diagnosed by a doctor?' Rhinitis was defined as sneezing, nasal congestion or rhinitis other than with respiratory infections, accompanied by itching of the eye and tearing during the previous 12 months. Age of onset of rhinitis was indicated by the parents at the time of doctor diagnosis. Eczema was defined with a positive answer to the question, 'Has your child ever had atopic eczema?'

Sociodemographic and perinatal characteristics

Information on the child's sex, maternal age, maternal educational level and the number of siblings was recorded in a structured questionnaire completed by the parents after delivery. Information on duration of gestation, birth weight and length, and maternal smoking during pregnancy was received from the Medical Birth Registries of Oulu and Tampere University Hospitals.

Statistical analysis

Cox proportional hazards regression was used to analyze the association between the antioxidants and asthma and rhinitis endpoints, whereas logistic regression was used for the eczema endpoint. The standardized Z-scores of total and dietary maternal antioxidant intake in their continuous forms were used, which were then adjusted for total energy in the analysis. The assumption of linearity was fulfilled. For supplemental antioxidants, we dichotomized maternal intake into consumers and non-consumers without adjustment for total energy. None of the women took supplemental α -carotene, α - and γ -tocopherols, whereas only 0.2% took retinol; therefore, these supplements were not studied. Multiple hazards and logistic regression models were computed, adjusting for the following variables: sex of child, place of birth (Southern or Northern Finland), season of birth (winter, spring, summer, autumn), gestational age at birth (in quartiles), maternal age at birth (<25 years, 25–29 years, 30–34 years, $\geq$ 35 years), maternal basic education (less than high school, high school graduate), maternal smoking during pregnancy (no, yes, no information), mode of delivery (vaginal, forceps or suction, cesarean section), number of siblings (none, one, two or more), parental asthma (no, yes, no information), parental allergic rhinitis (no, yes, no information), pets at home at 1 year of age (no, yes), and atopic eczema by 6 months of age (no, yes). Additional models were computed by adjusting for maternal intake of vitamin D and polyunsaturated fatty acids, and stratifying the analysis by season of birth and parental

history of allergy. All analyses were performed by using SPSS for windows (SPSS Inc., Chicago, IL, USA).

Results

Basic characteristics

The prevalence of asthma, rhinitis and eczema in the study population at the age of 5 years was 6, 14 and 37%, respectively. The intake of antioxidants during pregnancy is presented in Table 1. The mean intakes from supplements covered only a small fraction of the total intakes varying from 0.2% (retinol) to 23% (riboflavin). α -Carotene, α - and γ -tocopherols were not taken as supplements by the women during pregnancy. Participants in the allergy study were generally not systematically different from non-participants with regards to the background characteristics of the study population (Table 2).

Association between maternal intake of antioxidants and allergic outcomes

After adjusting for the potential confounders, maternal intake of any of the food-based antioxidants was not significantly associated with the risk of asthma, rhinitis or eczema in the offspring, except for the intake of magnesium,

Table 1 Maternal intake of antioxidants during pregnancy ($N = 2441$)

Antioxidants	From foods mean (s.d.)	From supplements	
		Proportion of consumers (%)	Mean (s.d.)
Total energy (KJ)	11 386 (3148)	N/A	N/A
Vitamin A (μ g)	1244 (755)	6.7	19 (95)
α -carotene	1398 (1587)	— ^a	— ^a
β -carotene	4258 (3508)	6.6	204 (903)
Vitamin C (mg)	194 (111)	34	25 (88)
Vitamin E (mg)	11 (3.9)	37	1.2 (4.3)
α -tocopherol	11 (3.9)	— ^a	— ^a
γ -tocopherol	10 (4.5)	— ^a	— ^a
Zinc (mg)	17 (4.7)	27	2.2 (4.4)
Copper (mg)	1.9 (0.6)	5.8	0.1 (0.3)
Selenium (μ g)	85 (25)	17	7.0 (14)
Magnesium (mg)	474 (127)	29	20 (51)
Calcium (mg)	1853 (686)	20	62 (171)
Manganese (mg)	6.7 (2.5)	26	0.4 (0.8)
Folate (μ g)	364 (113)	39	48 (92)
Riboflavin (mg)	2.9 (1.0)	41	0.9 (2.4)
Retinol (μ g)	861 (669)	0.2	1.8 (59)

^aSupplemental α -carotene, α -tocopherol, and γ -tocopherol were not consumed by the mothers during pregnancy.

N/A: Supplements did not contribute to total energy intake of the mothers.

Table 2 Distribution of the background characteristics by participation in the allergy study

Characteristic	All (N = 3523) n (%)	Participants (n = 3253) n (%)	Non-participants (n = 270) n (%)	P-value
<i>Place of residence</i>				<0.001
Northern Finland	1509 (43)	1451 (96)	58 (4)	
Southern Finland	2014 (57)	1802 (90)	212 (10)	
<i>Season of birth</i>				0.894
Winter	917 (26)	852 (93)	65 (7)	
Spring	870 (25)	802 (92)	68 (8)	
Summer	1436 (41)	1322 (92)	114 (8)	
Autumn	300 (9)	277 (92)	23 (8)	
<i>Duration of gestation</i>				0.651
1. quartile: ≤38.99 weeks	875 (25)	811 (93)	64 (7)	
2. quartile: 39.00–39.99 weeks	837 (24)	767 (92)	70 (8)	
3. quartile: 40.00–40.70 weeks	869 (25)	797 (92)	72 (8)	
4. quartile: >40.70 weeks	909 (26)	848 (93)	61 (7)	
Missing	33 (1)	30 (91)	3 (9)	
<i>Maternal age</i>				0.663
<25 years	573 (16)	526 (92)	47 (8)	
25–29 years	1229 (35)	1131 (92)	98 (8)	
30–34 years	1030 (29)	960 (93)	70 (7)	
≥35 years	691 (20)	636 (92)	55 (8)	
<i>Maternal basic education</i>				0.636
Less than high school	1241 (35)	1145 (92)	96 (8)	
High school graduate	2160 (61)	1998 (93)	162 (7)	
Missing	122 (4)	110 (90)	12 (10)	
<i>Maternal smoking during pregnancy</i>				0.502
No	3117 (89)	2875 (92)	242 (8)	
Yes	40 (1)	35 (88)	5 (13)	
No information	304 (9)	285 (94)	19 (6)	
Missing	62 (2)	58 (94)	4 (6)	
<i>Mode of delivery</i>				0.946
Vaginal delivery	2798 (79)	2580 (92)	218 (8)	
Forceps or suction	212 (6)	196 (93)	16 (7)	
Cesarean section	485 (14)	451 (93)	34 (7)	
Missing	28 (1)	26 (93)	2 (7)	
<i>Number of siblings</i>				0.131
None	1488 (42)	1378 (93)	110 (7)	
One	1117 (32)	1018 (91)	99 (9)	
Two or more	877 (25)	821 (94)	56 (6)	
Missing	41 (1)	36 (88)	5 (12)	

which was independently associated with a reduced risk of eczema (OR: 0.78; 95% CI: 0.62–0.97). Furthermore, maternal intake of vitamin C, folate and riboflavin from foods was appreciably associated with eczema only before adjustment (Table 3). Maternal total intake of any of the antioxidants was not significantly associated with any of the endpoints (data not shown).

Compared with non-consumers, consumers of supplemental vitamin C, zinc, selenium, magnesium and manganese were more likely to have children with increased risk of eczema, but these were only significant before adjustment. Similarly, maternal intake of supplemental vitamin C,

copper and calcium was positively related to rhinitis only before adjustment (data not shown).

Further adjustment for maternal intake of vitamin D and polyunsaturated fatty acids did not alter any of these results. Stratification of the results by month of birth or by parental history of allergy (parental rhinitis or asthma) did not alter the results. When we redefined the asthma outcome by studying doctor-diagnosed asthma (excluding wheezing symptoms or use of asthma medication during the preceding 12 months), our results were also similar. Finally, similar results were observed when wheezing alone was studied as an outcome (any wheezing symptom during the preceding 12 months).

Table 3 Association between maternal dietary antioxidant intakes during pregnancy and allergic endpoints in the offspring

Antioxidant	Hazard ratio (95% confidence interval) for asthma		Hazard ratio (95% confidence interval) for allergic rhinitis		Odds ratio (95% confidence interval) for atopic eczema	
	Unadjusted	Adjusted	Unadjusted	Adjusted	Unadjusted	Adjusted
Vitamin A	1.22 (0.98–1.53)	1.20 (0.92–1.56)	1.06 (0.94–1.19)	1.05 (0.92–1.19)	1.06 (0.96–1.17)	1.12 (0.99–1.26)
α -carotene	1.18 (0.93–1.48)	1.18 (0.86–1.62)	1.08 (0.98–1.19)	1.07 (0.97–1.18)	1.07 (0.98–1.17)	1.06 (0.95–1.18)
β -carotene	1.15 (0.91–1.46)	1.15 (0.84–1.58)	1.09 (0.99–1.20)	1.07 (0.97–1.19)	1.07 (0.97–1.16)	1.04 (0.93–1.17)
Vitamin C	0.98 (0.78–1.23)	0.92 (0.70–1.22)	0.94 (0.82–1.07)	0.93 (0.82–1.07)	0.90 (0.81–0.99) ^a	0.91 (0.80–1.03)
Vitamin E	0.98 (0.73–1.30)	1.02 (0.70–1.47)	0.96 (0.81–1.14)	0.99 (0.83–1.18)	0.97 (0.85–1.10)	1.00 (0.84–1.19)
α -tocopherol	0.98 (0.73–1.30)	1.02 (0.70–1.47)	0.96 (0.81–1.14)	0.99 (0.83–1.18)	0.97 (0.85–1.10)	1.00 (0.84–1.19)
γ -tocopherol	1.10 (0.88–1.37)	1.01 (0.76–1.34)	0.92 (0.80–1.05)	0.95 (0.83–1.10)	1.02 (0.92–1.13)	1.07 (0.93–1.22)
Zinc	1.04 (0.73–1.48)	1.15 (0.77–1.72)	1.09 (0.88–1.35)	1.08 (0.86–1.35)	0.90 (0.76–1.06)	0.92 (0.74–1.15)
Copper	0.96 (0.72–1.29)	1.00 (0.71–1.41)	1.11 (0.92–1.33)	1.08 (0.89–1.31)	0.99 (0.86–1.15)	0.94 (0.78–1.14)
Selenium	1.31 (0.91–1.89)	1.37 (0.89–2.11)	0.99 (0.81–1.20)	0.99 (0.80–1.22)	0.89 (0.76–1.04)	0.99 (0.81–1.21)
Magnesium	0.90 (0.62–1.29)	0.91 (0.59–1.41)	0.97 (0.78–1.20)	0.95 (0.76–1.19)	0.81 (0.69–0.96) ^a	0.78 (0.62–0.97) ^a
Calcium	1.07 (0.85–1.33)	1.14 (0.89–1.48)	0.99 (0.85–1.14)	0.97 (0.83–1.14)	0.90 (0.80–1.02)	0.96 (0.83–1.11)
Manganese	0.96 (0.79–1.18)	0.94 (0.73–1.20)	1.00 (0.88–1.14)	1.00 (0.87–1.15)	0.95 (0.86–1.05)	0.87 (0.76–1.00)
Folate	1.04 (0.77–1.42)	1.04 (0.71–1.52)	0.96 (0.80–1.15)	0.95 (0.79–1.15)	0.87 (0.75–1.00) ^a	0.91 (0.75–1.09)
Riboflavin	1.03 (0.82–1.31)	1.07 (0.83–1.40)	0.93 (0.79–1.09)	0.89 (0.75–1.05)	0.87 (0.76–0.98) ^a	0.87 (0.74–1.03)
Retinol	1.17 (0.95–1.45)	1.16 (0.91–1.50)	1.01 (0.90–1.14)	1.01 (0.89–1.15)	1.03 (0.94–1.13)	1.10 (0.98–1.24)

^a $P < 0.05$.

Discussion

Generally, the results from this study did not indicate significant evidence for the association between intake of total, dietary or supplemental antioxidants during pregnancy and the risk of asthma, rhinitis and eczema in the offspring when adjustment was made for potential confounders, except for the intake of dietary magnesium, which was associated with a decreased risk of eczema. Further adjustment for the effect of maternal vitamin D and polyunsaturated fatty acids intake and stratification by parental allergies and month of birth of the child did not alter these results.

Prenatal antioxidant exposures may program the child's susceptibility to allergic disorders already *in utero* (Prescott *et al.*, 1999; Devereux, 2007). Our results did not confirm the protective effects that have been observed between the intake of vitamin E and zinc and the risk of asthma (Devereux *et al.*, 2006). Even when we redefined the asthma outcome or when we studied wheezing symptom in the preceding 12 months as an outcome, the results were similar. Akin to previous studies, we observed no association between vitamin E and the risk of eczema. However, because of the poor validity of the FFQ for vitamin E (Erkkola *et al.*, 2001), we consider the data partly insufficient to rule out a possible protection of maternal vitamin E intake against asthma and wheezing. In the validation study, the overall quintile notation was, however, acceptable for both zinc and vitamin E: 73 and 65%, respectively, were correctly classified in the same or adjacent quintile by both methods (Erkkola *et al.*, 2001). In contrast to previous reports, we did not find any significant effect of supplemental folate on asthma

(Håberg *et al.*, 2009; Whitrow *et al.*, 2009). Rather, we observed a potential unadjusted protective effect of maternal intake of dietary folate on the risk of eczema. In addition, we did not confirm the associations that have been reported between vitamin C (Litonjua *et al.*, 2006), calcium (Miyake *et al.*, 2010b) and iron (Shahen *et al.*, 2004) and the risk of eczema. So far, no study has investigated the effect of maternal intake of antioxidants on the risk of rhinitis in the offspring. Nonetheless, our results showed no significant association between any of the antioxidants and the incidence of rhinitis.

The inconsistent results across studies, despite similar methodologies in the assessment of maternal intake of antioxidants and the outcomes, may probably be explained by two scenarios. It is possible that the differences in the amount of maternal intake of antioxidants across studies may explain the discrepancies in findings. For instance, the mean daily intake of vitamins C, vitamin E and zinc was 108.4, 7.8, and 7.3 mg, respectively, in a Japanese study (Miyake *et al.*, 2010a), whereas the mean daily intake of these nutrients was 279.6, 30.7 and 30.3 mg in a US study (Litonjua *et al.*, 2006). The corresponding figures from a UK study were 119, 8.2 and 12.5 mg (Devereux *et al.*, 2006). In the present study, the total mean daily intake of vitamin C was 221.4 mg, vitamin E 12.3 mg and zinc 18.8 mg. The discrepancies also indicate that there may be a critical time window when maternal exposure to antioxidants might be essential in influencing the risks of allergies in the offspring. The timing of assessment of maternal intake of antioxidants during pregnancy has not been uniform across studies. For instance, although some studies assessed dietary intake during the first and second trimester (Litonjua *et al.*, 2006;

Håberg *et al.*, 2009; Whitrow *et al.*, 2009), others assessed during the second trimester only (Martindale *et al.*, 2005; Devereux *et al.*, 2006), still others asked about maternal dietary intake at anytime during pregnancy (Miyake *et al.*, 2010a, b). Maternal diet, although assessed during the eighth month of pregnancy in our study, was aimed at reflecting the overall picture of maternal diet during the first 8 months of pregnancy. In addition, the use of supplements was inquired for the whole pregnancy period in our study. These differences at the timing of assessment of dietary intake may imply a possibility of detecting different effects of maternal dietary intake on the development of allergies in the offspring. Therefore, it would be important to find out the most likely time window in pregnancy during which maternal intake of antioxidants may have greater effect on the development of allergic outcomes in the offspring.

Maternal intake of dietary magnesium was inversely associated with the risk of eczema in this study. Considering the number of tests performed, we cannot rule out that this observation may be a chance finding. This effect, however, was not altered after further adjustment for vitamin D and polyunsaturated fatty acids, and stratification by parental allergies, indicating a possible true effect. Magnesium may be beneficially related to hyperactivity and pulmonary functions (Bichara and Goldman, 2009) and may protect against the risk of asthma and allergies (Soutar *et al.*, 1997). Although the mechanism responsible for this action is not clearly understood, it is possible that, in addition to the maintenance of cellular homeostasis, magnesium participates in stabilizing T cells and inhibiting mast cell degranulation, thus reducing inflammatory mediators (Bichara and Goldman, 2009). A recent double-blind placebo-controlled trial reported a reduced bronchial reactivity to methacholine among 7- to 19-year-old children who were given oral magnesium supplementation for 2 months (Gontijo-Amara *et al.*, 2007).

The unadjusted associations between some of the supplements and allergies indicate that intake of supplements can act partly as proxy for other lifestyle characteristics or dietary patterns. This necessitates identifying broader behavioral patterns behind the use of dietary supplements during pregnancy. It has been shown that those belonging to older age groups, having longer education and normal weight before pregnancy may use supplements more frequently (Arkkola *et al.*, 2006). These are the same groups that tend to have higher intake of most antioxidants from dietary sources as well (Uusitalo *et al.*, 2008).

One of the strengths of this study lies in its prospective nature, so that the ascertainment of the exposure before the outcome gave us the possibility to estimate more closely the putative effects of the antioxidants on the outcomes, minimizing differential bias regarding the selection of the subjects. We utilized a validated FFQ in assessing maternal dietary intake, which has been shown suitable for measuring the intake of diet of pregnant Finnish women (Erkkola *et al.*, 2001). It is possible that the nonsignificant findings we

observed with most of the antioxidants may indicate the difficulty of the FFQ to accurately quantify maternal antioxidant intake during pregnancy. However, the validity estimates for the main dietary sources of antioxidants (Uusitalo *et al.*, 2008) were acceptable except for those of dietary oils (Erkkola *et al.*, 2001).

Our outcomes were assessed using the standard ISAAC questionnaire, modified to the Finnish context. Asthma was defined by combining the question on doctor-diagnosed asthma with the questions on wheezing symptoms or use of asthma medication during the preceding 12 months. As wheezing alone may not accurately predict asthma in the long term, we believe this combination enabled us to estimate the true asthma cases. In our validation of the asthma component of the ISAAC questionnaire, this combination gave 98% sensitivity and specificity (Nwaru *et al.*, 2010). Our subjects were carrying genetic risk for type 1 diabetes. Type 1 diabetes is associated with Th1 cells, whereas allergic disorders are related to Th2 cell types (Pralhalad, 2000). In addition to the complexities surrounding the various components in the immune system that relate to the production of Th1 and Th2 cells, it is not clear how type 1 diabetes and allergies may be associated (Stene and Joner, 2004; Dales *et al.*, 2005). It is, therefore, possible that our results may not be completely generalizable to the general population. The cumulative incidence of asthma, rhinitis and eczema, however, paralleled that observed from the general Finnish population (Haahtela *et al.*, 2008). Compared with those who did not participate in the allergy study, participants were generally not different, minimizing possible selection bias.

Conclusion

Maternal intake of dietary magnesium during pregnancy may protect against atopic eczema in the offspring. We did not confirm the observations reported in previous studies concerning other antioxidants. These inconsistencies may result from the variable amount of maternal intake of antioxidants across studies. It may also indicate that there may be a critical time window in pregnancy when maternal intake of antioxidants might modulate the risk of developing allergic disorders in the offspring.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgements

We are grateful to the Finnish type 1 diabetes prediction and prevention doctors, research nurses, nutritionists and laboratory staff for their continuous collaboration through

the years. We also express our gratitude to Sirpa Pohjola and Ilona Kalliomäki for their expert technical assistance. This work was supported by the following: The Academy of Finland (grants 44105, 48724, 80846, 201988, 126813, 129492); the Finnish Pediatric Research Foundation; Doctoral Programs in Public Health; the Juho Vainio Foundation; the Yrjö Jahnsson Foundation; Medical Research Funds, Turku; Oulu and Tampere University Hospitals; Juvenile Diabetes Research Foundation; Novo Nordisk Foundation; the University of Tampere Foundation; and EU Biomed 2 Program (BMH4-CT98-3314).

References

- Arkkola T, Uusitalo U, Pietikäinen M, Metsälä J, Kronber-Kippilä C, Erkkola M *et al.* (2006). Dietary intake and use of dietary supplements in relation to demographic variables among pregnant Finnish women. *Br J Nutr* **96**, 913–920.
- Asher MI, Keil U, Anderson HR, Beasley R, Crane J, Martinez F *et al.* (1995). International Study of Asthma and Allergies in Childhood (ISAAC): rationale and methods. *Eur Respir J* **8**, 483–491.
- Bichara MD, Goldman RD (2009). Magnesium for the treatment of asthma in children. *Can Fam Physician* **55**, 887–889.
- Dales R, Chen Y, Lin M, Karsh J (2005). The association between allergy and diabetes in the Canadian population: implications for the Th1-Th2 hypothesis. *Eur J Epidemiol* **20**, 713–717.
- Devereux G (2007). Early life events in asthma—diet. *Pediatr Pulmonol* **42**, 663–673.
- Devereux G, Turner SW, Craig LCA, McNeill G, Martindale S, Harbour PJ *et al.* (2006). Low vitamin E intake during pregnancy is associated with asthma in 5-year-old children. *Am J Respir Crit Care Med* **174**, 499–507.
- Erkkola M, Karppinen M, Javanainen J, Räsänen L, Knip M, Virtanen SM (2001). Validity and reproducibility of a food frequency questionnaire for pregnant Finnish women. *Am J Epidemiol* **154**, 466–476.
- Gontijo-Amara C, Ribeiro MAGO, Gontijo LSC, Condino-Neto A, Ribeiro JD (2007). Oral magnesium supplementation in asthmatic children: a double-blind randomized placebo-controlled trial. *Eur J Clin Nutr* **61**, 54–60.
- Haahtela T, von Hertzen L, Mäkelä M, Hannuksela M, Allergy Programme Working Group (2008). Finnish allergy programme 2008–2018—time to act and change the course. *Allergy* **63**, 634–645.
- Håberg SE, London SJ, Stigum H, Nafstad P, Nystad W (2009). Folic acid supplements in pregnancy and early childhood respiratory health. *Arch Dis Child* **94**, 180–184.
- Kay AB (2001). Allergy and allergic diseases: first of two parts. *N Engl J Med* **344**, 30–37.
- Kupila A, Muona P, Simell T, Arvilommi P, Savolainen H, Hämäläinen AM *et al.* (2001). Feasibility of genetic and immunological prediction of type I diabetes in a population-based birth cohort. *Diabetologia* **44**, 290–297.
- Li-Weber M, Giasisi M, Trieber MK, Krammer PH (2002). Vitamin E inhibits IL-4 gene expression in peripheral blood T-cells. *Eur J Immunol* **32**, 2401–2408.
- Litonjua A, Rifas-Shiman SL, Ly NP, Tantisira KG, Rich-Edwards JW, Camargo Jr CA *et al.* (2006). Maternal antioxidant intake in pregnancy and wheezing illnesses in children at 2 y of age. *Am J Clin Nutr* **84**, 903–911.
- Martindale S, McNeill G, Devereux G, Campbell D, Russell G, Seaton A (2005). Antioxidant intake in pregnancy in relation to wheeze and eczema in the first two years of life. *Am J Respir Crit Care Med* **171**, 121–128.
- Miyake Y, Sasaki S, Tanaka K, Hirota Y (2010a). Consumption of vegetables, fruit, and antioxidants during pregnancy and wheeze and eczema in infants. *Allergy* **65**, 758–765.
- Miyake Y, Sasaki S, Tanaka K, Hirota Y (2010b). Dairy food, calcium, and vitamin D intake in pregnancy and wheeze and eczema in infants. *Eur Respir J* **35**, 1228–1234.
- Nwaru BI, Lumia M, Kaila M, Luukkainen P, Tapanainen H, Erkkola M *et al.* (2010). Validation of the Finnish ISAAC questionnaire on asthma against anti-asthmatic medication reimbursement database in 5-year-old children. *Clin Respir J*; DOI: 10.1111/j.1752-699X.2010.00222.x.
- Prahalad S (2000). Atopy, autoimmunity, and the T(H)1/T(H)2 balance. *J Pediatr* **137**, 446–449.
- Prescott SL, Macaubas C, Smallacombe T, Holt BJ, Sly PD, Holt PG (1999). Development of allergen-specific T-cell memory in atopic and normal children. *Lancet* **353**, 196.
- Remes ST, Korppi M, Kajosaari M, Koivikko A, Soininen L, Pekkanen J (1998). Prevalence of allergic rhinitis and atopic dermatitis among children in four regions of Finland. *Allergy* **53**, 682–689.
- Seaton A, Godden DJ, Brown K (1994). Increase in asthma: a more toxic environment or a more susceptible population? *Thorax* **49**, 171–174.
- Shahen SO, Newson RB, Henderson AJ, Emmett PM, Sherriff A, Cooke M *et al.* (2004). Umbilical cord trace elements and minerals and risk of early childhood wheezing and eczema. *Eur Respir J* **24**, 292–297.
- Soutar A, Anthony A, Brown K (1997). Bronchial reactivity and dietary antioxidants. *Thorax* **52**, 166–170.
- Stene LC, Joner G, Norwegian Childhood Diabetes Study Group (2004). Atopic disorders and risk of childhood-onset type 1 diabetes in individuals. *Clin Exp Allergy* **34**, 201–206.
- Uusitalo L, Uusitalo U, Ovaskainen ML, Niinistö S, Kronberg-Kippilä C, Marjamäki L *et al.* (2008). Sociodemographic and lifestyle characteristics are associated with antioxidant intake and the consumption of their dietary sources during pregnancy. *Public Health Nutr* **11**, 1379–1388.
- Whitrow M, Moore VM, Rumbold AR, Davies MJ (2009). Effect of supplemental folic acid in pregnancy on childhood asthma: a prospective birth cohort study. *Am J Epidemiol* **170**, 1486–1493.
- Virtanen SM, Kenward MG, Erkkola M, Kautiainen S, Kronberg-Kippilä C, Haukilahti T *et al.* (2006). Age at introduction of new foods and advanced beta cell autoimmunity in young children with HLA-conferred susceptibility to type 1 diabetes. *Diabetologia* **49**, 1512–1521.