

Diet, wheeze, and atopy in school children in Menorca, Spain

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Epidemiological studies have shown inverse associations of asthma symptoms with fish, vegetable, and fruit intake. We evaluated the association between several dietary factors with wheeze and atopy among children in Menorca, a Spanish Mediterranean island. A cross-sectional analysis was performed on 460 children at age 6.5 yr. Parents completed a questionnaire on the child's respiratory and allergic symptoms, and a 96-item food frequency questionnaire. Children underwent skin prick tests with six common aeroallergens. The average daily intake was relatively high for fruits (177 g) and fish (54 g), and moderate for vegetables (59 g). A high consumption (> 40 g/day) of fruity vegetables (tomatoes, eggplants, cucumber, green beans, zucchini) was found to have beneficial effect on current wheeze [odds ratio (OR), 0.38; 95% confidence interval (CI), 0.15–0.95, $p < 0.05$], and atopic wheeze with a significant decreasing trend when intake was increased (OR, 0.19; 95% CI, 0.04–0.95, p for trend = 0.04). No other fruits or vegetables were significantly associated with wheeze or atopy prevalence. An inverse association was found between a fish intake ≥ 60 g/day and atopy (OR, 0.43; 95% CI, 0.21–0.90, $p < 0.05$). The associations remained significant after adjustment for energy intake and maternal diet during pregnancy. Our results support a potential protective effect of fruity vegetables and fish intake during childhood on wheeze and atopy respectively.

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Diet has been recently recognized as a potential risk factor for asthma and allergic disorders, although the epidemiological evidence to date is still conflicting (1–3). Fruits and vegetables are rich sources of antioxidant vitamins such as vitamins C, E and carotenoids, and other antioxidants such as selenium and flavonoids, that are thought to reduce airway inflammation by protecting airway cells from endogenous and exogenous oxidative damage (4). Fish is a major source of long chain n-3 polyunsaturated fatty acids (PUFAs) that may downregulate the immune responses towards the T helper 2 (Th2) cells and provide a preventive effect on the inflammatory response (5, 6).

Epidemiological studies in children have consistently shown beneficial effects of fruit and vegetables consumption on asthma and related outcomes (7–12), whereas for fish intake the

results have been inconsistent. Four studies reported a protective effect of fish consumption on asthma-related respiratory symptoms in children (11, 13–15), whereas two other studies found no association (8, 16).

In a previous analysis we found that maternal fish consumption during pregnancy had a protective effect on eczema, atopy, and atopic wheeze among children in Menorca (17). We used the data of the same cohort of women enrolled during pregnancy and whose children were followed up to 6.5 yr of age to test the hypothesis whether children's diet has an impact on asthma symptoms and atopy.

Methods

The study methods have been described in detail previously (18). Women presenting for antenatal

care at all general practices in Menorca ($n = 507$; 95% of eligible mothers) over a 12-month period starting in mid-1997 were recruited. Four hundred and eighty-two children were subsequently enrolled and 468 provided complete outcome data after 6.5 yr of follow-up. Eight children were excluded from the analysis due to implausible values for total energy intake (outside the range of 800–3000 kcal/day). The outcomes of interest were current wheeze (described as one or more episodes of 'whistling or wheezing from the chest, but not noisy breathing from the nose' over the last 12 months), atopic wheeze at age 6.5 yr (defined as current wheeze and atopy at this age), and atopy at 6.5 yr (based on skin prick test (SPT) response).

During the follow-up at 6.5 yr, parents answered a questionnaire reporting all medical events over the preceding 12 months, and other questions on socio-demographic and lifestyle variables. Mothers were interviewed at 6 months, 14 months, and 2 yr after delivery regarding the type and duration of exclusive breastfeeding. Child body mass index (BMI) at 6.5 yr was calculated from the weight and height measured with the same instrument in all subjects using standardized protocol. Weight and height measurements were available for 415 children (415, 90.2%). Children with a BMI at or above the age- and sex-specific 85th BMI percentile were classified as overweight, whereas those above the 95th BMI percentile were classified as obese (19, 20).

A total of 412 children (412, 89.6%) underwent skin prick testing at 6.5 yr of age using a series of six common aeroallergens: *Dermatophagoides pteronyssinus*, grass pollen, olea europea, mixed gramineae, parietaria, and cat epithelium. Histamine and saline solutions were used as positive and negative controls, respectively. A positive SPT was defined as the presence of a mean wheal diameter of 2 mm or greater than the response to the negative uncoated control (18). Selection of a more specific cut-off (3 mm) gave basically the same results. A child was considered atopic if he or she demonstrated a positive reaction to one or more of the six allergens.

A semi-quantitative food frequency questionnaire (FFQ) of 96 food items was used to assess usual dietary intake in children aged 6.5 yr. The FFQ was a modified version of the Harvard questionnaire (21) that was adapted for use, and previously validated, in other adult Spanish populations (22, 23). The original food list was modified to include foods frequently eaten by children, and serving sizes were also reduced conveniently to avoid potential overestimation of

intakes. Foods were grouped into eight broad categories: (i) dairy products; (ii) meat, poultry, and fish; (iii) vegetables and legumes, (iv) fruits and nuts; (v) breads and cereals; (vi) sweets; (vii) oils and fats; and (viii) beverages. Children parents in our study were asked how often, on average, their children had consumed each type item over the previous year. The questionnaire had nine possible responses, ranging from 'never or less than once per month' to 'six or more per day.' For each participant, the usual daily intake of each food and food groups was estimated in grams per day based in the standard portion sizes. Total energy intake was estimated using nutrient composition from the foods composition tables of the US Department of Agriculture (<http://www.nal.usda.gov/fnic/foodcomp>). Maternal dietary habits during pregnancy were measured with a shorter (some foods were regrouped into food groups) Spanish version of the validated European Prospective Investigation into Cancer and Nutrition-Norfolk (EPIC-Norfolk) FFQ (24).

Data analysis was performed using SPSS 13.0 (SPSS Inc, Chicago, IL, USA). As children with atopy and children with wheeze may differ, we created a four-category wheeze-atopy status variable (both wheeze and atopy, wheeze according to the questionnaire alone, atopy alone, and neither wheeze nor atopy). Vegetables and fruits were classified into sub-groups according to the classification used in EPIC study (25). Vegetable intake did not include consumption of potatoes, legumes, and vegetable juices.

Univariate logistic regression analysis was first used to test for differences between dietary variables (using the lowest tertile as reference) and the outcomes of interest (yes/no). Multivariate logistic regression models were further used to adjust for confounders and to test for interactions. The following variables were considered as potential confounding factors: gender, maternal and parental asthma, maternal and paternal atopy, maternal smoking, BMI at age 6.5 yr, maternal and paternal education and social class, breastfeeding, fish intake during pregnancy, and number of siblings at age 6.5 yr. In addition, analyses including specific food intake were adjusted for total energy intake using the standard multivariate method (26). All variables significantly related with the outcome of interest in the bivariate models ($p < 0.2$) were included in the multivariate model. Odds ratios (OR) and 95% confidence intervals (95% CI) were computed to estimate the degree of association. To test for linear trend (Wald test) food intake was included as an ordinal variable in the logistic model.

Table 1. Characteristics of the study population*

Exposure variable	n (%)
Sex	
Male	232 (50.4)
Female	228 (49.6)
Maternal smoking	
Never	295 (65.3)
Ex-smoker	27 (6.0)
Current smoker	130 (28.8)
Father's education	
Less than primary school	47 (10.4)
Primary school	250 (55.6)
Secondary school	115 (25.6)
University	38 (8.4)
Mother's education	
Less than primary school	33 (7.4)
Primary school	223 (50.1)
Secondary school	128 (28.8)
University	61 (13.7)
Maternal asthma	
No	428 (93.0)
Yes	32 (7.0)
Maternal atopy	
No	286 (64.0)
Yes	161 (36.0)
Breastfeeding	
No	79 (17.1)
Yes	382 (82.9)
BMI	
Normal	284 (68.4)
Overweight	73 (17.6)
Obese	58 (14.0)
Number of siblings	
0	88 (19.1)
1	275 (59.8)
≥2	97 (21.1)
Current wheeze at age 6.5 yr	
Yes	40 (8.7)
No	420 (91.3)
Atopy† at age 6.5 yr	
Yes	70 (17.0)
No	342 (83.0)
Atopic wheeze‡ at age 6.5 yr	
Yes	20 (5.8)
No	324 (94.2)

*Data are presented as n (%), which give the number of subjects with available information on the specific variable.

†At least one skin prick test positive.

‡The reference group included those children that had no current wheeze, neither atopy.

Results

The characteristics of the children who participated in the study are shown in Table 1. The prevalence of current wheeze, atopic wheeze, and atopy at age 6.5 yr were 8.7%, 5.8%, and 17.0%, respectively. Table 2 presents the average daily intakes (g/day) for fish, fruits, and vegetables as well as for the foods included in each food group. The main subgroup of vegetables was fruity vegetables accounting for about half of total vegetable intake (54%). Among fruity vegetables,

Table 2. Description of daily consumption (g/day) of fish, fruits, and vegetables

	Mean (s.e.)
<i>Fish</i>	54.09 (1.36)
Fatty fish	6.79 (0.33)
Fatty fish (tuna, swordfish, anchovy, salmon); griddle or boiled	1.99 (0.22)
Canned fish (tuna, sardine or mackerel in oil)	4.79 (0.23)
Non-fatty fish	15.12 (0.59)
Non-fatty fish (hake, sole, gilthead bream); griddle or boiled	11.00 (0.53)
Fish croquettes	4.12 (0.26)
Fried or coated fish	14.36 (0.59)
Seafood	6.62 (0.25)
Lam, mussel, cockle	1.38 (0.10)
Squid, baby squid, cuttlefish, octopus	3.21 (0.15)
Crab, lobster, prawn, king prawn	2.03 (0.12)
<i>Fruits</i>	176.70 (4.03)
Fruits with vitamin C (orange, tangerine, kiwi)	47.43 (2.69)
Fruits with β-carotene (peach, melon, apricot)	39.52 (1.46)
Berries (grapes, cherries, strawberries)	15.88 (0.58)
Apples	54.93 (1.99)
Bananas	35.92 (1.29)
<i>Vegetables</i>	58.55 (1.89)
Leafy vegetables (lettuce, green salad, spinach)	13.72 (0.63)
Fruity vegetables	31.38 (1.17)
Tomatoes	18.29 (0.92)
Green beans	9.75 (0.50)
Eggplant, zucchini, cucumber	2.88 (0.20)
Root vegetables (grated-cooked carrots, beetroot)	10.12 (0.53)
Cabbages (cabbage, cauliflower, broccoli)	3.33 (0.33)

Table 3. Prevalence rates of current wheeze and atopy per daily intake of fish, fruits, and vegetables in children of Menorca

Food groups	Tertiles (g/day)	Mean intake (g/day)	Current wheeze (n = 40)	Atopy† (n = 70)	Atopic wheeze‡ (n = 20)
Fish	≤38.7 38.7–60.5 >60.5	25.4 48.5 87.3	11.18 7.89 7.05	19.85 21.28 10.00*	6.80 6.90 4.00
Fruits	≤138.3 138.3–210.3 >210.3	87.4 170.6 269.0	7.95 8.55 9.55	21.09 13.14 17.01	7.62 3.45 6.50
Vegetables	≤36.2 36.2–73.6 >73.6	16.9 54.8 102.6	9.21 9.87 7.05	17.52 18.12 15.33	6.14 6.96 4.35
Fruity vegetables§	≤17.1 17.1–40 >40	7.2 29.3 58.7	10.63 10.88 4.58	22.70 14.29 13.77	8.85 6.96 1.72*

*p < 0.05 with first tertile as reference.

†At least one skin prick test positive.

‡The reference group included those children that had no current wheeze, neither atopy.

§Fruity vegetables: tomatoes, eggplants, cucumber, green beans, zucchini.

the most consumed were tomatoes (58%) and green beans (31%). Apple was the most commonly eaten fruit, accounting for approximately 31% of total fruit consumption.

Table 3 presents the crude prevalence of current wheeze, atopy, and atopic wheeze per tertile

of intake of fish, fruits, vegetables, and fruity vegetables. A high intake of fish was inversely associated with the prevalence of atopy, whereas a high intake of fruity vegetables (tomatoes, eggplants, cucumber, green beans, zucchini) was inversely associated with atopic wheeze. For the other sub-groups of vegetables (root, leafy vegetables, cabbages), and the different sub-groups of fruits (citrus, berry fruits, other fresh fruits) we did not observe significant associations with wheeze or atopy. Similarly, no significant associations were observed with other food groups (diary products, cereals, legumes, meat and poultry, sweets and sugar, lipids) tested (data not shown).

Table 4 presents the associations of current wheeze, atopy, and atopic wheeze with the daily intakes of fish, fruits, vegetables, and fruity vegetables after adjusting for potential confounders. A high consumption (>40 g/day) of fruity vegetables was found to have beneficial effect on current wheeze (OR, 0.38; 95% CI, 0.15–0.95, $p < 0.05$), and atopic wheeze with a significant decreasing trend with increasing intake (OR, 0.19; 95% CI, 0.04–0.95, p for trend = 0.04). Atopy was negatively associated with a high fish intake (>60.5 g) (OR, 0.43; 95% CI, 0.21–0.90, $p < 0.05$). When we simultaneously included fruity vegetables and fish intake in the multivariate models, results remained very similar, showing an independent beneficial effect on the prevalence of atopy and wheeze (data not shown).

Discussion

The present study showed that children in Menorca had a considerable intake of fish, fruits and fruity vegetables, and that the intakes of fish and fruity vegetables had a beneficial effect on atopy and current wheeze respectively.

Fruity vegetables (tomatoes, eggplants, cucumber, green beans, zucchini) contain many potentially important antioxidants that cannot easily be quantified. A study on plasma carotenoid levels in relation to fruit and vegetable intake showed that fruity vegetables were associated with plasma α -carotene levels, while intake of tomatoes was significantly positively associated with plasma lycopene, and also with β -carotene levels (27). When we studied separately intake of tomatoes (fresh tomatoes, tomato sauce) we did not identify beneficial associations with wheeze or atopy, probably due to lack of variability in tomato intake in the study population, whereas a high consumption of green beans (≥ 35.6 g/day) showed a protective effect on current wheeze though not statistically significant (OR, 0.18; 95% CI, 0.02–1.44, $p = 0.102$). Green beans are known to be high sources of vitamin C and vitamin A (through their concentration of β -carotene). The Third National Health and Nutrition Examination Survey (NHANES III) showed that high levels of α -carotene and vitamin C in serum were the only antioxidants levels significantly associated with a lower risk for asthma among children in USA (28). Although other studies on nutrition and asthma in children have observed a beneficial effect of fruit intake on asthma symptoms, we did not observe any significant associations in the present study.

Fish, in particular oil fish, is one of the most important sources of dietary long-chain n-3 PUFAs (3). The n-3 PUFAs eicosapentaenoic (EPA) and docosahexaenoic (DHA) found in fish oil competitively inhibit the formation of prostaglandins and leukotrienes derived from arachidonic acid, and also have a suppressive effect on neutrophil function and thus an anti-inflammatory potential (6, 29). The mechanisms by which they may prevent atopic sensitization are unknown.

Table 4. Association (odds ratios and 95% CI*) of current wheeze and atopy with the daily intake of fish, fruits and vegetables among children of Menorca

	Current wheeze† (n = 40)			Atopy‡ (n = 70)			Atopic wheeze § (n = 20)		
	Tertiles of daily intake		p for trend	Tertiles of daily intake		p for trend	Tertiles of daily intake		p for trend
	T2	T3		T2	T3		T2	T3	
Fish	0.60 (0.28–1.33)	0.48 (0.21–1.09)	0.075	1.16 (0.63–2.13)	0.43 (0.21–0.90)	0.033	1.26 (0.42–3.79)	0.56 (0.16–2.00)	0.338
Fruits	0.91 (0.37–2.25)	0.89 (0.38–2.11)	0.793	0.60 (0.31–1.17)	0.90 (0.48–1.70)	0.741	0.48 (0.134–1.73)	0.98 (0.32–2.99)	0.980
Vegetables	0.97 (0.43–2.11)	0.61 (0.26–1.43)	0.255	1.09 (0.58–2.05)	0.94 (0.48–1.83)	0.860	1.19 (0.40–3.55)	0.69 (0.19–2.46)	0.558
Fruity vegetables	0.92 (0.44–1.94)	0.38 (0.15–0.95)	0.044	0.61 (0.32–1.16)	0.60 (0.31–1.15)	0.106	0.78 (0.28–2.20)	0.19 (0.04–0.95)	0.043

*Adjusted for: †gender, maternal asthma, and total energy intake; ‡gender, maternal asthma, maternal atopy, fish intake during pregnancy, and total energy intake; and §gender, maternal asthma, maternal atopy, BMI, fish intake during pregnancy, and total energy intake. The reference group for each food item includes those subjects in the first tertile of consumption.

There are only a few observational studies among children that evaluated the association between atopy and fish intake, and their results are inconsistent (30–32). A case-control study in Finland observed no association between fish intake and atopy at baseline, but after 9 yr the children who developed an atopic disease consumed less fish at baseline (32). The Childhood Allergy and Respiratory Health Study showed a significant association between fish intake and ryegrass sensitization (OR, 0.37; 95% CI, 0.15–0.90), but not with house dust mite sensitization (OR, 0.87; 95% CI, 0.36–2.13) (30). Another study showed that wheeze ever, doctor visits for wheeze, bronchodilator use, and nocturnal coughing were significantly reduced in children with high levels of ω -3 fatty acids in their plasma at age 18 months (33). We have previously shown that maternal fish intake during pregnancy had a protective effect on the risk of eczema at 1 yr of age, the risk of atopy, and atopic wheeze at 6.5 yr of age (17). We tested for interaction between maternal fish intake during pregnancy and children's fish intake at age 6.5 yr and found no significant interactions ($p = 0.508$). The results from both analyses are consistent and indicate that a high fish intake during pregnancy and childhood may provide protection on the risk of atopic sensitization.

One strength of the present study is that unlike previous epidemiological studies on diet and asthma in children, food intake was adjusted by total energy which allowed us to control for unknown confounders and better address whether dietary reporting might have been biased, e.g., by identifying energy under-over-reporters (3). Furthermore, the study population included children from the follow-up of a birth cohort, giving us the opportunity to account for the effect of early life exposures (such as maternal diet during pregnancy, breastfeeding) measured prospectively within the cohort. This is the first time to our knowledge that a study assessed simultaneously maternal dietary habits during pregnancy and children dietary habits in relation to asthma and atopy outcomes in childhood.

Still the cross-sectional nature of our analysis limits the interpretation of our results, as it is not optimal for the assessment of causal relationships. A high intake of fish and vegetables may reflect a more healthy diet and even a healthier lifestyle in general. However, when we adjusted for other health-related lifestyle and social factors such as maternal smoking during pregnancy and childhood, parental education and social class, we did not observe significant confounding effect. Finally, we have considered

parental reports on children's diet and symptoms, and information bias could have occurred. Studies of nutrition in childhood suggest that food frequency methods produce both valid and reproducible estimates of the dietary intakes of children and adolescents (34, 35). Moreover, wheeze at age 6.5 yr is less associated with low respiratory infections than at an earlier age and therefore more likely to be a marker of asthma.

In conclusion, the current study showed a potential protective effect of fruity vegetables and fish intake on childhood wheeze and atopy, respectively. The biological mechanisms underlying the observed associations need to be further investigated.

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