

Original article

Dietary intake in sensitized children with recurrent wheeze and healthy controls: a nested case–control study

Background: The rising prevalence of asthma and allergic disease remains unexplained. Several risk factors have been implicated including diet, in particular poly-unsaturated fats and antioxidant intake.

Methods: A nested case–control study comparing the dietary intake of sensitized children with recurrent wheeze (age 3–5 years) and nonsensitized children who had never wheezed was carried out within an unselected population-based cohort. Cases and controls were matched for age, sex, parental atopy, indoor allergen exposure and pet ownership. Dietary intake was assessed using a validated semi-quantitative food frequency questionnaire and nutrient analysis program.

Results: Thirty-seven case–control pairs (23 male, mean age 4.4 years) participated. Daily total polyunsaturated fat intake was significantly higher in sensitized wheezers (g/day, geometric mean, 95% confidence intervals: 7.1, 6.4–7.9) compared with nonsensitized nonwheezy children (5.6, 5.0–6.3, $P = 0.003$). Daily omega-3 and omega-6 fat intakes were not significantly different between the two groups. No significant differences were found in intake of any antioxidant or antioxidant cofactors between the groups.

Conclusions: Young sensitized wheezy children had a significantly higher total polyunsaturated fat intake compared with nonsensitized nonwheezy children. However, we were unable to distinguish a significant difference in specific polyunsaturated fat intakes. Otherwise the children in both groups had a very similar nutritional intake.

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The rapid increase in the prevalence of asthma and atopic disease is likely because of changes in lifestyle and/or in the environment. Amongst other factors, nutrition may have played a role. Two hypotheses have been put forward regarding different aspects of diet (1): (1) reduction in dietary antioxidants (vitamin A, C and E) and/or antioxidant cofactors (selenium, copper and zinc) consequent to reduction in intake of fresh fruit and vegetables and an increase in convenience foods; (2) shift in fatty acid intake from omega-3 to omega-6 polyunsaturated fats because of an increase in margarine consumption and decline in animal fat consumption (2, 3).

We aimed to investigate whether there are any differences in dietary macro- and micro-nutrient intake between sensitized children with wheeze and nonsensitized asymptomatic children. We used a case–control study nested within the context of a prospective birth cohort, which allowed us to control for a number of risk factors, which have previously been associated with asthma and sensitization (parental sensitisation status and history of allergic disease, indoor allergen exposure, pet exposure, gender).

Methods

Study population

Within an unselected population-based birth cohort study (Manchester Asthma and Allergy Study), we carried out a nested case–control study comparing dietary intake of wheezy sensitized and nonwheezy nonsensitized children.

The cohort is described in detail elsewhere (4, 5). Participants were recruited prenatally, and attended review clinic at age 3 years (± 4 weeks). A standard respiratory questionnaire was interviewer-administered to collect the information on symptoms. Allergic sensitization (mite, cat, dog, grasses, milk and egg) was ascertained by skin prick testing; sensitization was defined as wheal at least 3 mm greater than the negative control. Indoor allergen levels were measured in dust samples collected at birth. Following the 3-year follow-up, parents were invited to enter their children into the case–control study. Based on the follow-up data, cases were defined using the following criteria:

- Sensitized (skin test positive to at least one allergen).
- Recurrent wheeze (at least three parentally-reported wheezy episodes).

Cases were matched with control children (skin test negative, no history of wheeze) according to gender, month of birth,

pet ownership, indoor allergen exposure and parental sensitization.

Dietary intake questionnaire

Following the selection, parents completed a semi-quantitative food questionnaire regarding current dietary intake. Nutrient intake was then calculated using DIETQ, a nutrient intake analysis program (Tinuviel Software, Warrington, UK), and the sixth Her Majesty's Stationery Office Edition of UK Composition of Foods. Further analysis of fatty acid intake was calculated using The Institute of Brain Chemistry UK Fatty Acid database.

Statistics

Analysis was carried out using SPSS version 10. The study has 80% power to detect a 24% or more difference between cases and controls assuming a discordant pair rate of 32%. To adjust for multiple testing we only accepted *P*-values of ≤ 0.01 as significant. We analysed demographic and nutrient data using the appropriate parametric (if the data or log-transformed data were normally distributed) or nonparametric tests. Further analysis was carried out using analysis of variance (ANOVA) adjusting for total energy or total fat intake.

Results

Of 541 children reviewed at age 3 years, 43 were skin test positive and had a history of at least three episodes of parentally reported wheeze. Of these, 37 parents agreed to take part in this study. Dietary intake questionnaires were completed on 37 (23 male) matched pairs.

Demographics

The mean age of the study population was 4.4 years. There were no significant differences between cases and controls in height [mean (95% CI); 104.0 cm (102.4–105.6) *vs* 104.7 cm (103.2–106.2), *P* = 0.51] or weight [17.9 kg (17.0–18.8) *vs* 18.2 (17.4–19.0), *P* = 0.59]. Seventeen children in each group had a cat, dog or both. There were no significant differences in the number of children born vaginally (26 *vs* 30, *P* = 0.28); maternal smokers (8 *vs* 4, *P* = 0.35) or the number who had older siblings (22 *vs* 19, *P* = 0.64, cases *vs* controls).

Macronutrients

There was no significant difference in mean daily energy intake between cases and controls or in mean daily protein, fat or carbohydrate intake (Table 1). Although total fat intake was not significantly different, polyunsaturated fat intake was significantly higher in cases compared with controls (Table 2). This remained significantly higher when adjusted for total fat intake [g/day, adjusted GM (95% CI): 7.0 (6.4–7.6) *vs* 5.7 (5.2–6.2), cases *vs* controls; mean difference 1.2 g/day (1.1–1.4), *P* = 0.001].

Table 1. Daily mean (95% CI) macronutrient intake in cases and controls, *P*-value paired *t*-test

	Case	Control	<i>P</i> -value
Energy (kcal)	1794 (1641, 1946)	1725 (1587, 1863)	0.52
Protein (g)	70.0 (63.4, 76.6)	66.4 (61.2, 71.7)	0.44
Total fat (g)	64.5 (58.3, 70.7)	60.3 (54.7, 65.9)	0.33
Carbohydrate (g)	248.9 (226.4, 271.4)	244.3 (223.6, 265.0)	0.77

Table 2. Mean (95% CI) daily fat intake in cases and controls, *P*-value paired *t*-test

	Case	Control	<i>P</i> -value
Saturated Fat (g)	30.1 (26.7, 33.6)	28.9 (26.1, 31.8)	0.58
Cholesterol (mg)	209.2 (183.2, 235.3)	200.4 (176.7, 224.0)	0.64
Monounsaturated fat (g)	21.1 (19.0, 23.1)	19.3 (17.3, 21.3)	0.23
Polyunsaturated fat (g)*	7.1 (6.4, 7.9)	5.6 (5.0, 6.3)	0.003

*GM (95% CI).

Vitamin and minerals

There were no significant differences in mean daily vitamin or mineral intakes between cases and controls (Table 3). There was a trend which failed to reach statistical significance for vitamin E intake to be higher in the cases compared with controls which remained nonsignificant after adjusting for total energy intake [adjusted GM (95% CI) mg/day, 5.7 (5.1–6.3) *vs* 4.9 (4.4–5.5), cases and controls, mean difference 1.2 (1.0–1.3), *P* = 0.06].

Fatty acids intake

There was a trend for mean daily intakes of omega-6 fatty acids to be higher in cases than controls but this failed to reach significance [mg/day, GM (95% CI): 4241.6 (3552.1–5065.5) *vs* 3557.5 (2957.5–4279.5), *P* = 0.16]. There were no significant differences in total mean daily intakes of omega-3 fatty acids between cases and controls [mg/day, GM (95% CI): 842.5 (666.9–1064.3) *vs* 712.5 (578.2–900.5), *P* = 0.33]. Differences in mean daily total omega-6 and omega-3 fatty acid intake between the groups remained nonsignificant after adjusting for total

Table 3. Mean (95% CI) daily intake of minerals and vitamins in cases and controls *P*-value paired *t*-test

	Case	Control	<i>P</i> -value
Retinol (µg)*	264.0 (160.5)	264.0 (128.0)	0.79
Carotene (µg)	2794.2 (2376.8, 3211.6)	2374.5 (2014.8, 2734.2)	0.15
Vitamin E (mg)†	5.7 (5.0, 6.6)	4.9 (4.4, 5.4)	0.06
Vitamin C (mg)†	121.0 (104.2, 140.6)	116.3 (96.02, 140.7)	0.74
Copper (mg)	0.72 (0.65, 0.78)	0.69 (0.63, 0.76)	0.66
Zinc (mg)	7.8 (7.0, 8.5)	7.4 (6.8, 7.9)	0.39
Selenium (µg)†	46.7 (41.6, 52.4)	45.4 (40.4, 51.0)	0.76

*Median (interquartile range), *P*-value Wilcoxon Signed Ranks test; †GM (95%CI).

Table 4. Adjusted geometric mean (95% CI) daily intakes of specific polyunsaturated fats (adjusted for total fatty acid intake) in cases and controls

	Case	Control	P-value
Total omega-3 fats (mg)	809.2 (686.8, 953.4)	751.4 (637.8, 888.8)	0.53
Total omega-6 fats (mg)	4105.2 (3608.3, 4670.4)	3677.5 (3229.2, 4183.9)	0.23
18:2 (n-6) Linoleic acid (mg)	3979.9 (3494.7, 4536.9)	3572.4 (3136.9, 4068.4)	0.25
20:4 (n-6) Arachidonic acid (mg)	70.0 (57.7, 84.7)	61.2 (50.5, 74.1)	0.33
18:3 (n-3) α -Linolenic acid (mg)	595.9 (492.7, 719.8)	555.6 (459.4, 671.8)	0.61
20:5 (n-3) Eicosapentaenoic acid (EPA) (mg)	43.9 (34.4, 56.2)	44.1 (34.5, 56.4)	0.98
22:6 (n-3) Docosahexaenoic acid (DHA) (mg)	82.4 (69.7, 97.2)	83.5 (70.7, 98.6)	0.91

fatty acid intake. There were no significant differences in mean daily intakes of individual omega-6 and omega-3 fatty acids between cases and controls (Table 4).

Discussion

In this case-control study of preschool sensitized children with recurrent wheeze and nonsensitized nonwheezy controls we have examined dietary intake using a semi-quantitative food frequency questionnaire. The only difference we found was that polyunsaturated fat intake was significantly higher in the symptomatic children compared with healthy controls. However, we could not find a significant difference in intake of any of the specific polyunsaturated fatty acids (PUFA). Intakes of the major omega-3 fatty acids [α -linolenic acid, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA)] were similar in cases and controls. However, there was a nonsignificant trend for intakes of the major omega-6 fatty acids (linoleic acid and arachidonic acid) to be higher in cases than controls.

We found no difference between dietary antioxidants (vitamin A, C and E) and/or antioxidant cofactors intake between cases and controls. Previous studies on this topic reported conflicting results. Vitamin C consumption has been associated with a reduction in wheezing and an increase in lung function in some (6, 7) but not all studies (8). Other studies have found an inverse relationship between vitamin E consumption and wheeze, but no relationship between vitamin C or A (9, 10).

The strength of our study is the very careful definition of the outcomes and matching of the cases and controls according to a number of factors which are associated with the development of atopic sensitization and wheeze. Furthermore, we used a semi-quantitative food frequency questionnaire along with a nutrient analysis program that allowed calculation of actual nutrient intake for each individual. The questionnaire and program have been validated in adults (11, 12). Portion sizes were altered accordingly for the use in young children. Although this method of calculating nutrient intake gives somewhat more information than many previous studies that have only inquired about types of foods eaten and not about frequency or quantity, semi-quantitative food frequency questionnaires have their limitations. They rely heavily on

recall and are only as good as the nutritional database that supports them. In addition they remain a 'snap-shot' of current diet, albeit a more overall picture than some other methods of assessing dietary intake (e.g. 48 h recall) used in some other studies (13).

The main weakness of our study is the relatively small number of participants. This may explain why we found the intake of PUFA to be significantly greater in cases than controls, but could not find a significant difference in individual omega-3 and omega-6 fatty acid intake.

Over the last few decades the type of fat consumed in western diets has changed with a reduction in saturated fats (animal fats) and an increase in PUFA in the form of vegetable-oil products rich in omega-6 fatty acids. The major omega-6 fatty acid consumed in the human diet is linoleic acid and this is converted to arachidonic acid. Human inflammatory and immune cells contain high proportions of arachidonic acid (14) and its principal functional role is as a substrate for the two-series prostaglandins (e.g. PGE₂) and the four-series leukotrienes. It has been suggested that this pattern of change in fat intake over recent decades is responsible for the corresponding increase in atopic disease (15). This is because PGE₂ exerts effects that could promote the development of a Th2-type phenotype. PGE₂ has been reported to increase the production from T-cells of IL-4 (16), IL-5 and IL-10 (Th2 cytokines) (17) and to decrease the production of IFN- γ and IL-2 (Th1 cytokines) (18).

As the consumption of vegetable oils (rich in omega-6 fatty acids) has increased, the consumption of omega-3 fatty acids has decreased both in absolute and relative (to omega-6 fatty acids) terms (19). For example, linoleic acid contributes 50–80% of fatty acids found in corn, sunflower and soybean oils, whereas good sources of α -linolenic acid such as rapeseed and soybean oils contain only 5–15% of these fatty acids. EPA and DHA are found in relatively high proportions in the tissues of 'oily fish', but in the absence of significant consumption of oily fish, α -linolenic acid is the major dietary omega-3 fatty acid. Since there is a competition between the omega-6 and omega-3 fatty acids for the enzymes that metabolise them, the relative reduction in omega-3 fatty acid intake may exacerbate the effect of the increase in omega-6 fatty acid intake. Thus, both relative and absolute increases in omega-3 fatty acid intake have the effect of reducing the amount of arachidonic acid produced.

A number of studies to date have found an association between margarine consumption and atopic disease. Margarine contains up to 20 times more linoleic acid than butter (20). Von Mutius et al. reported that the rising prevalence of hay fever and atopy in children in East Germany in the 1990s had coincided with, amongst other things, a change in diet, in particular a dramatic rise in the use of margarine over butter (21). Subsequently, other investigators in Europe have used dietary surveys to show an association between atopic disease in childhood and increased consumption of margarine over butter (22–24).

Several studies using food frequency questionnaires have shown an association between oily fish consumption and a reduction in asthma symptoms (24, 25) and also an association between the consumption of fish in the first year of life and a reduction in risk of developing asthma in early childhood (26). However, no studies in childhood so far have analysed actual nutrient intake. A study from Australia has shown that a high dietary intake of PUFA (information derived from enquiring about type, but not quantity of fat used) was associated with an increased risk of recent asthma in preschool children (27).

Although to date there is still little evidence suggesting a role of dietary fatty acids in the development of asthma

and allergies, randomized controlled trials have been initiated, with interesting early results (28). Supplementation of omega-3 fatty acid (with and without dust mite avoidance) significantly reduced wheeze in the first 18 months of life (27). At age 3 years there was no effect of dietary intervention on wheeze, but there was a significant reduction in cough in atopic children (29). Fish oil supplementation in pregnancy may reduce neonatal cytokine responses to dust mite, cat, ovalbumin and PHA (30). Although this study was not powered for clinical outcomes, the infants in the fish oil supplementation group were less likely to be sensitized to egg and had milder eczema.

The results of our study add further to the evidence that dietary intake of PUFA plays a role in the development of atopic wheeze in young children. Further studies in larger groups of children, using semi-quantitative food frequency questionnaire and nutrient analysis programs, may help to answer this question.

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References

- McKeever TM, Britton J. Diet and asthma. *Am J Respir Crit Care Med* 2004;**170**:725–729.
- Raper NR, Marston RM. Levels and sources of fat in the US food supply. In: Ip C, editor. *Dietary fat and cancer*. Alan R. Liss, New York, pp. 127–152.
- Roberts DCK. Dietary factors in the fall of coronary heart disease mortality. *Prostaglandin Leukot Essential Fatty Acids* 1991;**44**:97–101.
- Custovic A, Simpson BM, Murray CS, Lowe L, Woodcock A. The National Asthma Campaign Manchester Asthma and Allergy Study. *Pediatr Allergy Immunol* 2002;**13** (Suppl. 15): 32–37.
- Lowe L, Murray C, Custovic A, Simpson BM, Kissen P, Woodcock A. Specific airway resistance in three year old children. *Lancet* 2002;**359**:1904–1908.
- Forastiere F, Pistelli R, Sestini P, Fortes C, Renzoni E, Rusconi F et al. Consumption of fresh fruit rich in vitamin C and wheezing symptoms in children. *Thorax* 2000;**55**:283–288.
- Cook DG, Carey IM, Whincup PH, Papacosta O, Chirico S, Bruckdorfer KR et al. Effect of fresh fruit consumption on lung function and wheeze in children. *Thorax* 1997;**52**:628–633.
- Romieu I, Mannino DM, Redd SC, McGeehin MA. Dietary intake, physical activity, body mass index, and childhood asthma in the Third National Health And Nutrition Survey (NHANES III). *Pediatr Pulmonol* 2004;**38**:31–42.
- Hijazi N, Abalkhail B, Seaton A. Diet and childhood asthma in a society in transition: a study in urban and rural Saudi Arabia. *Thorax* 2000;**55**:775–779.
- Nagel G, Nieters A, Becker N, Linseisen J. The influence of the dietary intake of fatty acids and antioxidants on hay fever in adults. *Allergy* 2003;**58**:1277–1284.
- Burr ML, Fehily AM, Rogers S, Welsby E, King S, Sandham S. Diet and reinfarction trial (DART): design, recruitment, and compliance. *Eur Heart J* 1989;**10**:558–567.
- Fehily AM, Yarnell JW, Butland BK. Diet and ischaemic heart disease in the Caerphilly Study. *Hum Nutr Appl Nutr* 1987;**41**:319–326.
- Wijga AH, Smit HA, Kerkhof M, de Jongste JC, Gerritsen J, Neijens HJ et al. Association of consumption of products containing milk fat with reduced asthma risk in preschool children: the PIAMA birth cohort study. *Thorax* 2003;**58**:567–572.
- Yacoub B, Pala HS, Cortina-Borja M, Newsholme EA, Calder PC. Encapsulated fish oil enriched in α -tocopherol alters plasma phospholipid and mononuclear cell fatty acid composition but not mononuclear cell functions. *Eur J Clin Invest* 2000;**30**:260–274.
- Black PN, Sharp S. Dietary fat and asthma: is there a connection? *Eur Respir J* 1997;**10**:6–12.
- Naito Y, Endo H, Aria KI, Coffman RL, Arai N. Signal transduction in Th clones: target of differential modulation by PGE₂ may reside downstream of the PKC-dependent pathway. *Cytokine* 1996;**8**:346–356.
- Snijderwint FGM, Kalinski P, Wierenga EA, Bos JD, Kapsenberg ML. Prostaglandin E₂ differentially modulates cytokine secretion profiles of human T-helper lymphocytes. *J Immunol* 1993;**150**:5321–5329.
- Benbernou N, Esnault S, Shin HCK, Fekkar H, Guenounou M. Differential regulation of INF-gamma, IL-10 and inducible nitric oxide synthase in human T cells by cyclic AMP-dependent signal transduction. *Immunol* 1997;**91**:361–368.

19. Peat JK, Salome CM, Woolcock AJ. Factors associated with bronchoal hyperresponsiveness in Australian adults and children. *Eur Resp J* 1992;**5**:921–929.
20. Michels K, Sachs F. Trans fatty acids in European margarines. *N Engl J Med* 1995;**333**:130–132.
21. Von Mutius E, Weiland SK, Fritzsche C, Duhme H, Keil U. Increasing prevalence of hay fever and atopy among children in Leipzig, East Germany. *Lancet* 1998;**351**:862–866.
22. Bolte G, Frye C, Hoelscher B, Meyer I, Wjst M, Heinrich J. margarine consumption and allergy in children. *Am J Respir Crit Care Med* 2001;**165**:277–279.
23. Dunder T, Kuikka L, Turtinen J, Rasanen L, Uhari M. Diet, serum fatty acids, and atopic diseases in childhood. *Allergy* 2001;**56**:425–428.
24. Hodge L, Salome CM, Peat JK, Haby MM, Xuan W, Woolcock AJ. Consumption of oily fish and childhood asthma risk. *MJA* 1996;**164**:137–140.
25. Nafstad P, Nystad W, Magnus P, Jaakkola JJ. Asthma and allergic rhinitis at 4 years of age in relation to fish consumption in infancy. *J Asthma* 2003;**40**:343–348.
26. Haby MM, Peat JK, Marks GB, Woolcock AJ, Leeder SR. Asthma in preschool children: prevalence and risk factors. *Thorax* 2001;**56**:589–595.
27. Mhrshahi S, Peat JK, Marks GB, Mellis CM, Tovey ER, Webb K et al. for the CAPS team. Eighteen month outcomes of house dust mite avoidance and dietary fatty acid modification in the Childhood Asthma Prevention Study (CAPS). *J Allergy Clin Immunol* 2003;**111**:162–168.
28. Gore C, Custovic A. Can we prevent allergy? *Allergy* 2004;**59**:151–161.
29. Peat JK, Mhrshahi S, Kemp AS, Marks GB, Tovey ER, Webb K et al. for the Childhood Asthma Prevention Study Team. Three year outcomes of dietary fatty acid modification and house dust mite reduction in the Childhood Asthma Prevention Study. *J Allergy Clin Immunol* 2004;**114**:807–813.
30. Dunstan JA, Mori TA, Barden A, Beilin LJ, Taylor AL, Holt PG et al. Fish oil supplementation in pregnancy modifies neonatal allergen-specific immune responses and clinical outcomes in infants at high risk of atopy: a randomised, controlled trial. *J Allergy Clin Immunol* 2003;**112**:1178–1184.