Postnatal Fish Oil Supplementation in High-Risk Infants to Prevent Allergy: Randomized Controlled Trial

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KEY WORDS
Fish oil supplementation, infants, omega 3 PUFA, allergy, eczema, allergy prevention

ABBREVIATIONS
AA—arachidonic acid
DHA—docosahexaenoic acid
EPA—eicosapentaenoic acid
LC—long chain
LT—leukotriene
n-3 PUFA—omega 3 polyunsaturated fatty acids
n-6 PUFA—omega 6 polyunsaturated fatty acids
PG—prostaglandin
SPT—skin prick test

WHAT'S KNOWN ON THIS SUBJECT: Declining dietary omega 3 polyunsaturated fatty acids has been associated with rising allergy prevalence and fish oil is therefore of interest in allergy prevention. Supplementation during pregnancy, but not after the age of 6 months, has achieved some allergy reductions.

WHAT THIS STUDY ADDS: We assessed the effect of fish oil supplementation from birth to 6 months, which has not been investigated previously. Our results, together with previous findings, will likely help define a "window of opportunity" for allergy intervention using fish oil supplements.

abstract

BACKGROUND AND OBJECTIVE: Relative deficiency of dietary omega 3 polyunsaturated fatty acids (n-3 PUFA) has been implicated in the rising allergy prevalence in Westernized countries. Fish oil supplementation may provide an intervention strategy for primary allergy prevention. The objective of this study was to assess the effect of fish oil n-3 PUFA supplementation from birth to 6 months of age on infant allergic disease.

METHODS: In a double-blind randomized controlled trial, 420 infants at high atopic risk received a daily supplement of fish oil containing 280 mg docosahexaenoic acid and 110 mg eicosapentaenoic acid or a control (olive oil), from birth to age 6 months. PUFA levels were measured in 6-month-old infants' erythrocytes and plasma and their mothers' breast milk. Eczema, food allergy, asthma and sensitization were assessed in 323 infants for whom clinical follow-up was completed at 12 months of age.

RESULTS: At 6 months of age, infant docosahexaenoic acid and eicosapentaenoic acid levels were significantly higher (both $P < .05$) and erythrocyte arachidonic acid levels were lower ($P = .003$) in the fish oil group. Although n-3 PUFA levels at 6 months were associated with lower risk of eczema ($P = .033$) and recurrent wheeze ($P = .027$), the association with eczema was not significant after multiple comparisons and there was no effect of the intervention per se on the primary study outcomes. Specifically, between-group comparisons revealed no differences in the occurrence of allergic outcomes including sensitization, eczema, asthma, or food allergy.

CONCLUSIONS: Postnatal fish oil supplementation improved infant n-3 status but did not prevent childhood allergic disease. *Pediatrics* 2012;130:1–9

The epidemic rise in allergic diseases has been linked with Western lifestyle changes, and although the specific environmental drivers are likely multifactorial, dietary changes are among the most likely candidates. Of these, a declining intake of omega 3 polyunsaturated fatty acids (n-3 PUFA) and concurrent rise in omega 6 polyunsaturated fatty acids (n-6 PUFA) in Western diets has been of key interest.[1,2] The n-6–derived arachidonic acid (AA) may have some antiinflammatory properties but, importantly, is the direct precursor of highly inflammatory eicosanoids, leukotriene (LT)B_4, and prostaglandin (PG)E_2, which can promote Th2 differentiation and IgE production.[3,4] By contrast, the n-3 PUFAs give rise to considerably less inflammatory eicosanoid derivates (PGE$_3$ and LTB$_5$) and, resolvins with inflammation-resolving potential[5,6] and may also influence cell membrane structure, cell signaling and antigen presentation (reviewed in Calder[7]). Given the overall antiinflammatory effects of n-3 PUFA, they may be important in restoring the potentially allergy protective qualities of traditional diets.

The consequences of relative n-3 PUFA deficiency are likely to be greatest early in life when patterns of immune responses are still developing. Of concern, dietary changes are reflected in the declining n-3 PUFA content in breast milk[8] with implications for n-3 PUFA status of developing infants.

In the clinical context, the majority of studies have explored the role of n-3 PUFA status in pregnancy. Both observational[9–15] and interventional [16–18] studies have suggested a protective relationship between maternal n-3 PUFA consumption in pregnancy and infant allergic outcomes (reviewed in Klemens et al[19]). In contrast, the only study to examine the potential role of postnatal fish oil supplementation (after the age of 6 months) of term infants found no effect on reducing allergic outcomes.[20–22]

Here, in a randomized controlled trial, we investigate the effects of fish oil from birth until 6 months of age on allergic outcomes in children at high allergic risk. Our aims were to (1) to determine whether fish oil supplementation in the early postnatal period can reduce the risk of subsequent allergic disease and (2) examine the relationship between PUFA levels at 6 months of age and the risk of subsequent allergic outcomes.

METHODS

The study design and methodology including power calculations have been described in detail elsewhere.[23] A brief summary follows.

Study Population

Healthy term infants of 420 allergic women in Perth, Western Australia, were recruited in a double-blind, randomized controlled study. Allergic (otherwise healthy), nonsmoking women with restricted fish intake were recruited at 36 weeks of pregnancy between January 6, 2005, and January 10, 2008, from private and public metropolitan antenatal clinics. Maternal allergy was defined by a positive skin prick test (SPT) and a history of allergic disease. The participants were made aware of the study hypothesis. Ethical approval was obtained from ethics committees of participating hospitals and informed written consent was obtained from mothers.

Intervention

Infants were randomized to receive either a daily supplement of fish oil supplement or olive oil as the control. Supplementation commenced at birth and ceased at 6 months of age. The fish oil capsules contained 650 mg fish oil and provided 280 mg docosahexaenoic acid (DHA) and 110 mg eicosapentaenoic acid (EPA). Placebo capsules contained 650 mg olive oil (66.6% n-9 oleic acid) (both from Ocean Nutrition, Ltd, Mulgrave, Nova Scotia, Canada). The fatty acid composition of the capsules (purchased in one batch in 2005) remained unchanged over the course of the study, and peroxide and acid levels remained compliant with the Australian standards. Because the trial took longer than anticipated and because the Ocean Nutrition product was discontinued, the final 27 children received similar capsules of fish oil (250 mg DHA and 60 mg EPA) or olive oil kindly provided by ν-Mega Ingredients Pty Ltd, Eight Mile Plains, Queensland, Australia. This brand substitution was endorsed and supervised by the Ethics Committee at Princess Margaret Hospital. Capsules were image and scent matched. At the completion of the intervention period, there was no significant difference between erythrocyte DHA or EPA ($P = .732$ and $P = .069$, respectively) or plasma phospholipid DHA or EPA ($P = .160$ and $P = .121$, respectively) levels between the 2 groups of participants receiving fish oil capsules from different suppliers, so all children were included in the final analysis.

Although the primary outcomes were analyzed on an "intention-to-treat" basis, data were also examined allowing for differences in capsule consumption between the groups (see later). Capsule consumption/adherence was based on capsule diaries and a count of the returned capsules, in addition to the infant fatty acid analysis (see later).

Randomization was completed by external staff via computer software using an unpredictable allocation sequence, stratified according to maternal and paternal atopic history and parity. Mothers and study personnel were unaware of the group allocation. It was recommended that capsules be administered to infants in the morning immediately before to the first daily breast or bottle

feed by piercing the capsule and squirting the oil into the infant's mouth.

Blood Collection and Processing

Peripheral blood from 6-month-old infants was obtained when mothers consented and when practically possible by venipuncture. Blood was collected into heparinized tubes and processed immediately after the clinic visit. Plasma was stored at −80°C and erythrocytes were washed before lipids were extracted with methanol and chloroform (2:1) and stored at −20°C until analysis in one batch.

Erythrocyte and Plasma Phospholipid Fatty Acid Analysis

Erythrocyte and plasma phospholipid fatty acids were analyzed as previously described.[24]

Breast Milk Fatty Acid Analysis

Maternal breast milk samples (up to 6 ml in 3 aliquots) were collected when infants were 3 and 6 months of age. Samples were immediately frozen and stored at −80°C for subsequent analysis as described in.[25]

Clinical Outcomes and Allergy Definitions

Infants were evaluated at 6 and 12 months of age with a detailed clinical history and examination. A child was classified as having "allergic disease" if he or she had a physician diagnosis of IgE-mediated food allergy, eczema, or asthma at these assessments. Information on respiratory symptoms (recurrent wheeze, asthma, and rhinitis) was also collected at 6 and 12 months, but the limitations are recognized at this age. The diagnostic criteria conformed to the published clinical guidelines[26] and a diagnosis of eczema was made in infants with typical skin lesions.[27] The extent and severity of the eczema were determined by use of the standardized SCORAD severity index.[27] IgE-mediated food allergy was defined

as a history of immediate symptoms (typically within 60 minutes) after contact with and/or ingestion of a food and a positive SPT response to the implicated food.

Allergen Skin Prick Testing

Allergic sensitization was assessed by SPT at the 12-month visit using common allergen extracts (milk, peanut, house dust mite, cat, grass, mold; Hollister-Stier Laboratories, Spokane, WA) and whole egg, as well as histamine as a positive control and glycerine as a negative control. A wheal diameter of ≥2 mm was considered positive.

Statistical Methods

The primary clinical outcomes were assessed on an "intention-to-treat" basis. Logistic regression was used to determine the odds ratios for specific allergic outcome according to group allocation. For continuous data, differences between the groups were determined by independent t test for normally distributed data and by Mann-Whitney U tests when data were not normally distributed. Linear regression was used to determine the relationship between feeding and supplementation variables and fatty acid status. All statistical analysis was performed by using SPSS software (Version 16 for PC, SPSS Inc, Chicago, IL). A P value of <.05 was considered statistical significant for all analysis.

RESULTS

Population Characteristics

A total of 420 infants were randomized: 218 to the fish oil group and 202 to the control group. Their characteristics are given on Table 1. Fig 1 illustrates the participant flow through the trial. After randomization, 62 participants withdrew previous to completing the supplementation period and/or did not wish to attend clinical visits. The withdrawal rate was significantly higher in the fish oil group (18.3% compared with 10.9% from the placebo group, $P =$

TABLE 1 Characteristics of the Complete Recruited Population ($n = 420$)

Characteristics	Placebo Group	Fish Oil Group	P
Participants recruited, (% of total)	202 (48.1)	218 (51.9)	
Antenatal characteristics			
Maternal family history of allergy, (% of group)	202 (100)	218 (100)	
Paternal allergy, (% of group)	101 (50)	113 (51.8)	.845
Maternal age, y[a]	33.2 ± 4.2	32.5 ± 4.8	.131
Paternal age, y[a]	35.6 ± 4.8	35.2 ± 5.6	.377
Parity, (% of previous children)[b]	0.71 ± 0.9	0.8 ± 0.9	.282
Pets in the home, (% of group)	128 (63.4)	125 (57.3)	.685
Birth characteristics:			
Gestation, wk[a]	39.4 ± 1.2	39.2 ± 1.13	.020*
Gender, (% males)	104 (51.5)	103 (47.2)	.430
Vaginal delivery, (% of group)	130 (64.4)	136 (62.4)	.686
Birth weight, g[a]	3509 ± 433	3443 ± 451	.133
Birth length, cm[a]	50.6 ± 2.3	50.2 ± 2.8	.059
Head circumference, cm[a]	35 ± 1.8	34.9 ± 1.8	.530
Season of birth			.720
Supplementation adherence and withdrawal rates			
Capsule adherence, %[b]	64.52 ± 27.2	56.7 ± 32.1	.056
Participants withdrawn by 12-mo visit, (% of group)	22 (10.9)	40 (18.3)	.031*
Participants not attending 12-mo visit, (% of group)	12 (5.9)	22 (10.1)	.119
Total participants not seen at 12-mo visit, (% of group)	34 (16.8)	62 (28.4)	.005**
Participants attended 12-mo visit and included in analysis, (% of group)	167 (51.7)	156 (48.3)	.626

* $P < .05$, ** $P < .01$, differences reaching statistically significance. Continuous data shown as mean ± SD.
[a] Normally distributed continuous data analyzed using t tests.
[b] Continuous data not normally distributed and analyzed using Mann-Whitney U tests.
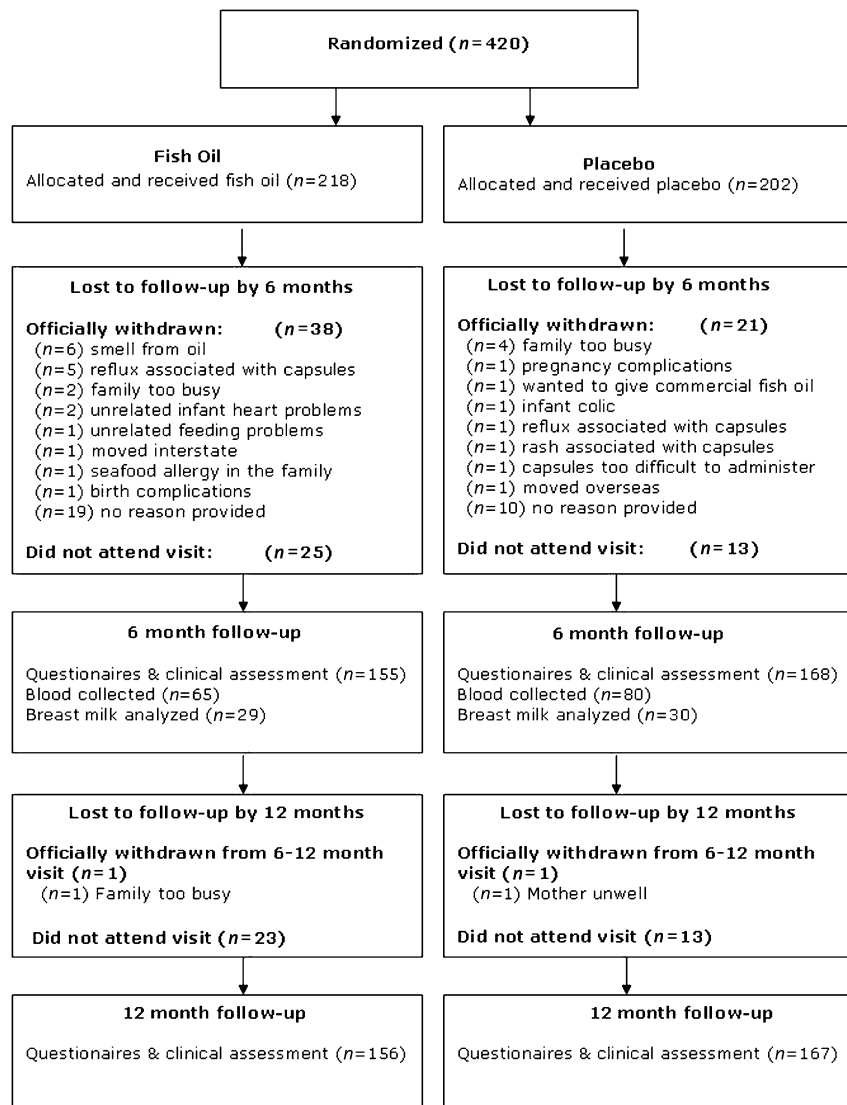
FIGURE 1
Flowchart outlining participant progression and data collected at 6- and 12-month visits.

shorter gestation ($P = .049$), the average gestation for both groups was in the 39th week and there were no differences in birth weight, length, or head circumference between the groups.

Effects of Infant Oil Supplementation on Infant PUFA Levels at 6 Months

At the end of the intervention, erythrocyte levels of DHA ($P = .03$) and EPA ($P = .016$) were significantly higher in the fish oil group compared with the control group (Fig 2A). AA levels were significantly lower in the fish oil group ($P = .003$), and oleic acid levels were not different between the study groups.

Plasma DHA ($P = .001$) and EPA ($P = .001$) levels were significantly higher in the fish oil group compared with in the control group (Fig 2B), but there were no differences in AA levels. Oleic acid levels were significantly higher in the placebo group ($P = .012$).

There were no differences in the PUFA status of these groups at birth (in cord blood erythrocytes) before the intervention (data not shown).

Factors Determining Fatty Acid Status at 6 Months of Age

In a subset of infants with measures of breast milk fatty acids, breast milk DHA content was correlated with erythrocyte and plasma DHA levels in the fish oil group (correlation coefficient 0.55 [$P = .008$] and 0.489 [$P = .003$], respectively) and in the control group (correlation coefficient 0.585 [$P = .001$] and 0.371 [$P = .043$], respectively). In all infants (fish oil and placebo groups combined), the adjusted r^2 values for the association between breast milk DHA and infant erythrocyte and plasma DHA were 0.292 and 0.157, respectively. In addition, breast milk DHA was the strongest predictor of erythrocyte DHA composition ($\beta = .527$, 95% confidence interval [CI] 1.442–4.882, $P = .001$), whereas plasma DHA was more strongly

.031), and participants in the fish oil group were more likely to withdraw because of the fishy smell. After the supplementation period, an additional number of participants could not be contacted or did not attend visits (Fig 1). Despite the use of vanilla flavoring in both oils, 92.2% of participants in the fish oil group correctly guessed their allocation, compared with 56.25% of the placebo group. Differences in supplementation adherence between the groups approached significance ($P = .056$) (Table 1), and this was assessed as a confounding factor.

The characteristics of the infants who attended the 12-month follow-up evaluation for allergic disease ($n = 323$) are given in Table 2. There were no differences in prerandomization characteristics of the participants who were lost to follow-up compared with those who were retained in the study apart from parental age, with younger parents more likely to discontinue the study (data not shown). In the follow-up population there were no major differences in the perinatal characteristics of infants who received the fish oil compared with those who received the control oil (Table 2). Although infants in the fish oil group had

TABLE 2 Characteristics of Children Assessed at 12 months of Age (n = 323)

	Placebo Group	Fish Oil Group	P
Participants attending 12-mo follow-up visit (with allergic outcomes), (% of total followed up)	167 (51.7)	156 (48.3)	
Participants withdrawn by 12-mo follow-up visit, (% of total recruited)	22 (10.9)	40 (18.3)	.031*
Participants not attending 12-mo follow-up visit, (% of total recruited)	12 (5.9)	22 (10.1)	.119
Antenatal characteristics			
Maternal family history of allergy, (% of group)	167 (100)	156 (100)	
Paternal allergy, (% of group)	114 (68.7)	111 (71.6)	.626
Maternal age, y[a]	33.6 ± 4.16	33.2 ± 4.44	.510
Paternal age, y[a]	35.9 ± 4.8	36.0 ± 5.5	.939
Parity[b]	0.77 ± 0.92	0.8 ± 0.95	.737
Pets at home, (% of group)	99 (60)	91 (59.9)	.981
Birth characteristics			
Gestation, wk[a]	39.4 ± 1.2	39.1 ± 1.1	.049*
Vaginal delivery, (% of group)	103 (62)	95 (62.5)	.934
Birth weight, g[a]	3502 ± 437	3438 ± 425	.180
Birth length, cm[a]	50.5 ± 2.3	50.1 ± 2.9	.154
Head circumference, cm[a]	35 ± 1.9	34.9 ± 1.9	.564
Season of birth			.771
Infant characteristics			
Male infants, (% of group)	84 (50.3)	81 (51.9)	.771
Average compliance, %	66.36	61.26	.129
Ever breastfed, (% of group)	163 (97.6)	152 (97,4)	.992
Duration breastfeeding, mo[b]	6.4 ± 3.6	6.7 ± 3.5	.557
Still breastfeeding at 6 mo, (% of group)	118 (72.8)	110 (76.9)	.413
Age solid introduction, mo[a]	5.5 ± 0.8	5.6 ± 1.0	.268
Daycare/playgroup attendance by 12 mo, (% of group)	142 (85.2)	122 (78.4)	.143
Growth parameters at 12 mo			
Weight at 12 mo, kg[a]	10.2 ± 1.4	10.1 ± 1.3	.919
Head circumference at 12 mo, cm[a]	46.8 ± 3.8	47 ± 3.7	.661
Height at 12 mo, cm[a]	76.7 ± 3.5	76.2 ± 3.42	.207

* $P < .05$, differences reaching statistically significance. Continuous data shown as mean ± SD.

[a] Normally distributed continuous data analyzed using t tests.

[b] Continuous data not normally distributed and analyzed using Mann-Whitney U tests.

predicted by fish oil intervention (adherence) (β = .943, 95% CI 0.015–0.046, $P < .000$) than breast milk DHA composition (β = .342, 95% CI 0.627–3.358, $P = .005$). In this study, 97.2% of infants received some breast milk and 74.6% were still receiving breast milk at 6 months (Table 1). Breast milk levels of oleic acid, AA, and DHA did not differ between the supplementation groups (data not shown).

Of the infants in the fish oil group, 70.2% were exposed to formula, compared with 61.9% in the control group (no significant difference $P = .127$). Of those who received formula, 54.3% in the fish oil group compared with 43.9% in the control group received a formula containing long-chain polyunsaturated fatty acid (LCPUFA; no significant difference $P = .164$)

Clinical Outcomes at 12 Months Between Fish Oil and Placebo Groups

There were no differences in prevalence of allergic outcomes (any allergic disease, overall sensitization, specific sensitization, eczema, or food allergy) between infants in the fish oil and control groups at 12 months of age (Fig 3). None of the children had a diagnosis of asthma by 12 months of age. There were no significant differences in recurrent wheeze or persistent coughing between the study groups at 6 or 12 months. All relationships remained nonsignificant after correction for capsule adherence, maternal age, paternal allergic disease, parity, gestation, gender, and maternal n-3 PUFA intake during pregnancy.

In infants who received more than 75% of the intended supplementation (the highest adherence quartile, No. = 145), there was a significantly lower prevalence of eczema at 12 months in the fish oil group ($P = .041$) but no significant differences between supplementation groups in any other allergic outcome.

The Relationship Between n-3 PUFA Status at 6 Months and Allergic Outcomes

We assessed the relationship between fatty acids levels and allergic outcomes in the combined population. Infants with higher erythrocyte EPA composition ($P = .033$) and higher EPA:AA ratio ($P = .022$) as well as higher plasma DHA levels ($P = .047$) at 6 months of age were significantly less likely to develop eczema by 12 months (Table 3).

These associations remained significant after adjustment for gender, paternal allergy, gestation, parity, and breastfeeding but were not significant after the addition of group allocation to the regression model (Table 3). Higher levels of AA ($P = .004$) and total n-6 PUFA ($P = .005$) (data not shown) levels at 6 months were associated with increased symptoms of eczema (recurrent dry, itchy, red and scaly patches of skin) at 6 months of age.

Elevated plasma levels of DHA ($P = .027$) and total LC n-3 PUFA (EPA, docosapentaenoic acid [DPA], and DHA) ($P = .028$) at 6 months associated with a reduced risk of recurrent wheeze in the first 12 months of life, and these associations remained significant after adjustment for gender, paternal allergy, gestation, parity, breastfeeding, and group allocation (Table 4). No other significant associations were observed between LCPUFA levels at 6 months and allergic outcomes.

DISCUSSION

The purpose of this study was to further explore whether improving n-3 PUFA status through postnatal fish oil
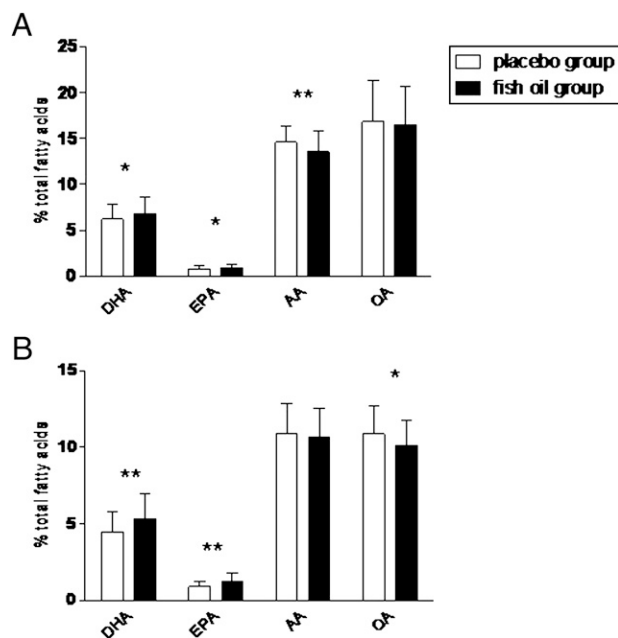
FIGURE 2
Infant fatty acid levels (% of total fatty acids) in (A) erythrocytes (*n* = 139) and (B) plasma (*n* = 145). Levels shown as mean ± SD. *P < .05 and **P < .001 indicate differences between the fish oil and placebo groups.

supplementation during early development can reduce the risk of allergic disease. Fish oil supplementation during pregnancy has achieved both immunomodulatory[16,28] and allergy protective [16–18,29] effects (systematically reviewed previously[19]), but no significant effects have been observed with postnatal supplementation after the age of 6 months.

Our results show that although the postnatal fish oil intervention was associated with potentially favorable effects on immune function at 6 months (D'Vaz et al, unpublished data), it did not achieve a reduction in the development of allergic disease in the first 12 months of life.

However, when examining the relationship between the n-3 PUFA levels at 6 months of age and allergic outcomes independent of supplementation group, we observed a protective relationship with subsequent eczema. This is consistent
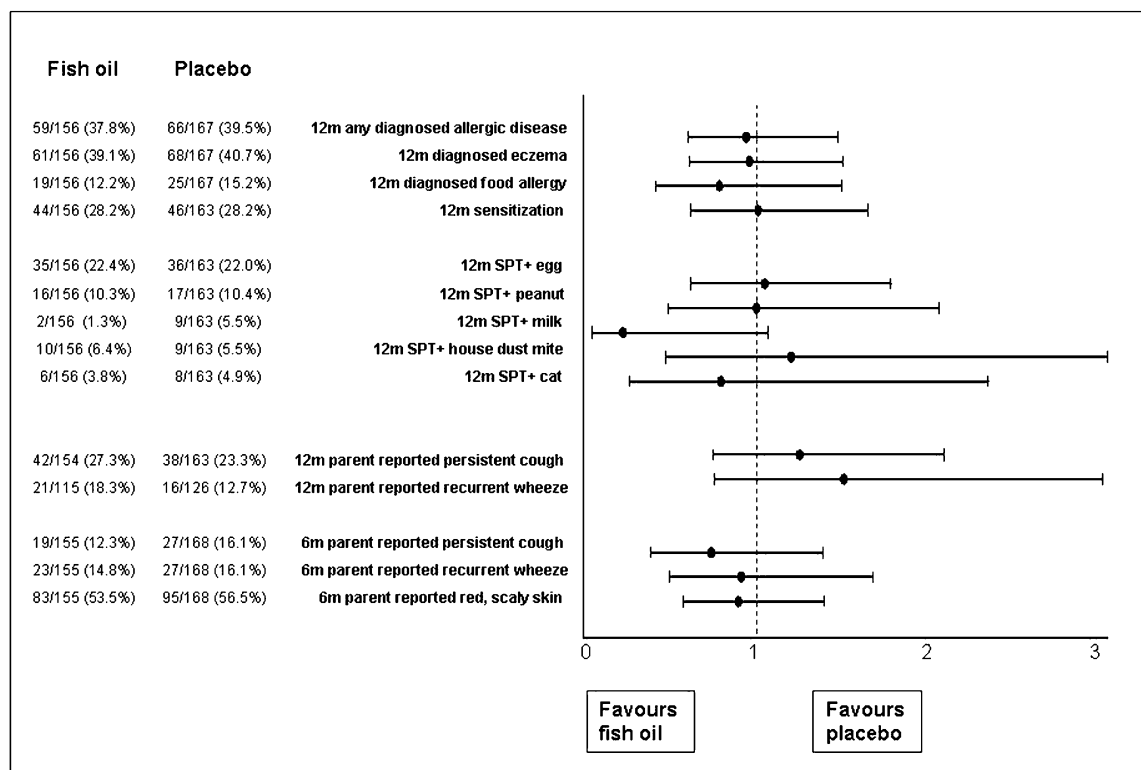

FIGURE 3
Left, Number and percentage of infants with allergic symptoms and disease at 6 and 12 months of age in the fish oil and placebo groups. Right, Odds ratios (mean, 95% CI) of children in the fish oil group having symptoms or disease compared with the placebo group.

TABLE 3 Comparison of Fatty Acid Levels Between Infants With and Without Eczema in the First Year of Life

	Infants With No Eczema	Infants With Eczema	Unadjusted Regression P	Adjusted[a] Regression P
Erythrocyte fatty acid levels at 6 mo (n = 139), % of total fatty acids, mean ± SD				
AA 20:4n6	14.0 ± 2.24	14.24 ± 1.78	.583	.638
EPA 20:5n3	0.91 ± 0.4	0.75 ± 0.4	.033*	.071
DHA 22:6n3	6.7 ± 1.7	6.35 ± 1.7	.236	.505
Total LC n3 (EPA + DPA + DHA)	9.5 ± 2.1	9.0 ± 2.1	.176	.325
EPA/AA	0.069 ± 0.04	0.054 ± 0.03	.022*	.055
Plasma fatty acid levels at 6 mo (n = 145), % of total fatty acids, mean ± SD				
AA 20:4n6	11.0 ± 1.8	10.7 ± 2.1	.401	.698
EPA 20:5n3	1.05 ± 0.48	1.01 ± 0.52	.639	.812
DHA 22:6n3	5.1 ± 1.42	4.6 ± 1.5	.047*	.103
Total LC n3 (EPA + DPA + DHA)	7.1 ± 1.88	6.51 ± 1.85	.063	.108
EPA/AA	0.1 ± 0.06	0.1 ± 0.058	.936	.867

* $P < .05$, differences reaching statistical significance. DPA, docosapentaenoic acid.
[a] Adjusted for gender, paternal allergy, gestation, parity, breastfeeding, and group allocation.

with other studies that have associated improved early n-3 PUFA status with reduction in the risk of IgE associated disease,[30] eczema,[17,31] and eczema severity.[16] Also in accordance with others,[32] plasma DHA levels were associated with reduced recurrent wheeze in the first year of life, indicating a possible effect on respiratory allergies. While Almquist et al[33] did not find an association between early fatty acid status and allergic disease at 5 years of age, we will further investigate these relationships at 2.5 and 5 years of age when more reliable measures of aeroallergen sensitization and asthma become available.

The association between n-3 PUFA levels and subsequent eczema, however, was nonsignificant after adjustment for group allocation, which may indicate that factors associated with the supplementation, other than the effect on

fatty acid levels, influenced the development of cutaneous symptoms.

So, while a relatively high n-3 status at 6 months was somewhat associated with reduced allergic outcomes, direct postnatal fish oil supplementation during very early infancy was not effective in preventing allergic disease, which is consistent with the lack of significant effects of fish oil supplementation previously observed in late infancy.[34,35] This, in conjunction with evidence from observational studies[9–12] and pregnancy supplementation studies may suggest that allergy protective effects of fish oil supplementation are more likely to occur earlier in development.

In one of the earliest pregnancy intervention studies, we observed immunomodulatory effects in neonates,[16,36–39] which was associated with reduced risk of allergen (egg) sensitization and eczema severity by 12 months of age.[16] Similar effects on immune function at birth[40] and infant clinical outcomes[17,31] of fish oil supplementation during pregnancy (some continuing through to lactation) have subsequently been confirmed by others. Additionally, a long-term beneficial effect of maternal fish oil supplementation on asthma development by 16 years of age was recently indicated.[18]

Immune studies show that significant differences in immune function are already evident at birth in children who develop allergic disease,[41,42] and these established patterns may be difficult to modify with a postnatal intervention. However, it is also possible that our intervention was ineffective due to variance in protocol adherence. In support of this, the protocol analysis revealed a significant reduction in eczema prevalence in the participants who were more than 75% compliant with the supplementation, suggesting that relatively high doses of fish oil in early infancy may be effective in reducing eczema prevalence.

TABLE 4 Comparison of Fatty Acid Levels Between Infants With and Without Recurrent Wheeze in the First Year of Life

	Infants With No History of Wheeze	Infants With Recurrent Wheeze	Unadjusted Regression P	Adjusted[a] Regression P
Erythrocyte fatty acid levels at 6 mo (n = 139), % of total fatty acids, mean ± SD				
AA 20:4n6	14.02 ± 2.04	14.52 ± 2.23	.419	.707
EPA 20:5n3	0.84 ± 0.43	0.96 ± 0.39	.360	.123
DHA 22:6n3	6.6 ± 1.66	5.97 ± 1.59	.208	.249
Total LC n3 (EPA + DPA + DHA)	9.29 ± 2.14	8.95 ± 1.73	.599	.761
EPA/AA	0.063 ± 0.037	0.07 ± 0.036	.527	.164
Plasma fatty acid levels at 6 mo (n = 145), % of total fatty acids, mean ± SD				
AA 20:4n6	10.88 ± 1.87	10.44 ± 2.47	.433	.374
EPA 20:5n3	1.06 ± 0.50	0.81 ± 0.28	.082	.220
DHA 22:6n3	5.01 ± 1.51	4.02 ± 1.09	.027*	.018*
Total LC n3 (EPA + DPA + DHA)	7.0 ± 1.92	5.76 ± 1.35	.028*	.029*
EPA/AA	0.10 ± 0.058	0.08 ± 0.039	.237	.511

* $P < .05$, differences reaching statistically significance. DPA, docosapentaenoic acid.
[a] Adjusted for gender, paternal allergy, gestation, parity, breastfeeding, and group allocation.

Despite the relatively high dose of n-3 PUFA used in our study, the increases in n-3 PUFA levels were modest, which could suggest issues with bioavailability and absorbtion of the ethyl ester supplements. Indeed, greater changes in infant fatty acid levels have been achieved by others using lower postnatal doses, delivered through maternal supplementation during lactation.[43] A study by Lauritzen et al also achieved relatively greater changes in infant fatty acid levels, which was interestingly not associated with allergic outcomes, although the study was not designed to evaluate allergy development.[44]

Finally, olive oil may not have been the optimum control oil, although it is commonly used as a placebo, because it has some immunomodulatory effects (see review[45]). Although n-9 oleic acid is found in high levels in breast milk and a relatively small additional amount was given in the placebo capsules, plasma oleic acid levels were higher in the placebo group. However, it is unlikely the olive oil obscured an effect of the fish oil as allergy rates in both groups are similar to those observed in our previous studies of high-risk infants (data not shown).

CONCLUSIONS

Fish oil supplementation from birth to 6 months modestly, but significantly, elevated n-3 LCPUFA levels and reduced n-6 AA levels at the end of the supplementation period, although breast milk LCPUFA levels were another major determinant of infant PUFA status at this age. Although higher n-3 LCPUFA levels were associated with reduced risk of subsequent allergic outcomes, this was not significant after adjustment for supplementation group and was only examined in a subgroup with fatty acid data measured. Importantly, the primary analysis revealed no significant effect of the intervention on infant allergic outcomes at 12 months of age. Thus, with emerging evidence of more allergy protective effects of LCPUFA in pregnancy compared with the postnatal period,[19,29] we suggest that optimizing n-3 PUFA status remains desirable but best achieved through promoting maternal n-3 PUFA intake during pregnancy possibly in conjunction with lactation.

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