Journal of Developmental Origins of Health and Disease

www.cambridge.org/doh

Human milk polyunsaturated fatty acids are related to neurodevelopmental, anthropometric, and allergic outcomes in early life: a systematic review

Saori Mitguard[1], Olivia Doucette[1] and John Miklavcic[1,2]

[1]Schmid College of Science and Technology, Chapman University, Orange, CA, USA and [2]School of Pharmacy, Chapman University, Irvine, CA, USA

Original Article

Cite this article: Mitguard S, Doucette O, and Miklavcic J. (2023) Human milk polyunsaturated fatty acids are related to neurodevelopmental, anthropometric, and allergic outcomes in early life: a systematic review. *Journal of Developmental Origins of Health and Disease* **14**: 763–772. doi: 10.1017/S2040174423000454

Received: 7 November 2023
Revised: 21 December 2023
Accepted: 24 December 2023
First published online: 23 January 2024

Keywords:
Neurodevelopment; allergy; growth; omega-3; omega-6

Corresponding author:
J. Miklavcic; Email: miklavcic@chapman.edu

Abstract

Polyunsaturated fatty acids are critically important for newborn nutrition and in the trajectory of growth and developmental processes throughout early life. This systematic review (PROSPERO ID: CRD42023400059) critically analyzes literature pertaining to how omega-3 and omega-6 fatty acids in human milk are related to health outcomes in early life. Literature selected for the review were published between 2005 and 2020 and included assessments in healthy term children between 0 and 5 years of age. The studies reported the relation between human milk fatty acids docosahexaenoic acid (C22:6*n*-3, DHA), eicosapentaenoic acid (C20:5*n*-3, EPA), alpha-linolenic acid (C18:3*n*-3, ALA), arachidonic acid (C20:4*n*-6, AA), and linoleic acid (C18:2*n*-6, LA) with three domains of health outcomes: neurodevelopment, body composition, and allergy, skin & eczema. Results from the 21 studies consistently suggested better health outcomes across the three domains for infants consuming milk with higher concentrations of total *n*-3, DHA, EPA, and ALA. Negative health outcomes across the three domains were associated with higher levels of total *n*-6, AA, and LA in milk. *N*-3 and *n*-6 content of milk were related to neurodevelopmental, body composition, and allergy, skin & eczema outcomes with *moderate* certainty. Maternal diet impacting milk fatty acid content and fatty acid desaturase genotype modifying physiologic responses to fatty acid intake were prominent gaps identified in the review using the NIH Quality Assessment Tool for Observational Cohort and Cross-Sectional Studies and GRADE approach. This research study can inform baby nutrition product development, and fatty acid intake recommendations or dietary interventions for mothers and children.

Introduction

N-3 and *n*-6 polyunsaturated fatty acids (PUFA) are critical constituents of lipid bilayer membrane composition. In addition to cellular structure, long chain PUFA are important bioactive compounds. The *n*-6 LA and *n*-3 ALA are essential fatty acids as they cannot be synthesized in humans. AA can be synthesized from LA, and DHA and EPA can be synthesized from ALA but at low rates and therefore should be supplied from diet.[1] Genetic variability in the fatty acid desaturase (FADS) gene cluster impacts metabolism of long chain PUFA as minor alleles in the *FADS* gene locus are associated with decreased DHA and AA levels.[2] Fatty acids in human milk are derived from endogenous synthesis in the mammary gland, maternal stores, uptake from plasma; and the presence of PUFA in milk depends on maternal dietary intake.[1] DHA level varies in human milk with maternal intake,[3,4] and maternal health status including obesity[5] and nonalcoholic fatty liver disease,[6] and there is debate regarding the optimal ratio of AA:DHA intake.[7] As a result, the dietary requirement for human milk PUFA in early life is poorly understood.

Tissue growth and composition of newborns are supported by fatty acids provided in human milk. *N*-3 fatty acid insufficiency has been associated with delayed or altered neural development.[8] Throughout the third trimester of pregnancy, accretion of DHA and AA in the fetus suggests the critical role of essential fatty acids in the coordination of typical growth and development. Furthermore, *n*-3 long chain PUFA are involved in the maintenance of immune function, neural plasticity, and synapse activity.[8] Differences in accretion rates of LA and ALA in the fetal cerebellum in the third trimester suggests unique contributions of *n*-3 and *n*-6 fatty acids to early development.[9] The body of research suggests important relations between the levels of *n*-3 and *n*-6 fatty acid intake and tissue levels with early life health outcomes.

There are very few studies relating the nutrient content of human milk to health outcomes in early life. For example, a review of human milk micronutrient content in relation to growth and body composition in the first two years only identified 28 studies over a 42 year period.[10]

There are even fewer studies of sufficient quality to inform dietary guidelines. Despite the importance of long chain PUFA in growth and development in early life, only three studies on fatty acid composition meeting the criteria to inform dietary reference intakes were identified in an evidence scan of published literature on milk nutrient content between 1980 and 2020.[11] Thus, the aim of this systematic review was to summarize how *n*-3 and *n*-6 fatty acids in human milk were related with neurodevelopment, anthropometric, and allergy outcomes in early life. A comprehensive review on this topic that explicitly makes considerations for fatty acid content (relative or absolute), control for covariates (diet, age, education, etc.), and genotype (*FADS*, *ELOVL*) in the assessment for certainty of evidence has not yet been published. This research is relevant and important because it can influence the direction of high-quality intervention research to inform dietary fatty acid intake recommendations in early life. The information can also inform formula and milk fortifier products that emulate human milk and provide a level of essential fatty acids that supports newborn growth and development.

Methods

The study is registered in PROSPERO (CRD42023400059)[12] and is reported on using the Preferred Reporting Items for Systematic Reviews and Meta-Analyses 2020 guidelines.[13]

Study identification and selection

Chapman University's Leatherby Library© catalog database, PubMed®, and Google Scholar© were used to find studies included in the review. To find literature that related health outcomes in early life to the milk content of *n*-3 and *n*-6 fatty acids, the search string was structured as: ("fatty acid" or "linoleic acid" or "alpha-linolenic acid" or "eicosapentaenoic acid" or "arachidonic acid" or "docosahexaenoic acid,") and ("newborn" or "neonat(e/al)" or "infant,") and ("body composition" or "growth" or "neurodevelopment" or "atopy" or "allergy" or "eczema,") and ("breast milk" or "breastfeeding" or "breast-feeding" or "infant feeding" or "lactation" or "colostrum" or "human milk.") To conduct the preliminary exclusion of initial search results, screening of titles was performed to eliminate reviews, animal or experimental research studies, studies with designs other than case-control or cohort/cross-section, pre-term children, description of fatty acid content only with no relation to health outcome, or the article was published before 2005 or after 2020. In the secondary screening of 122 search results, observational studies were selected only if they included healthy mothers, term babies from 0 to 5 years of age, an English copy was available, and the PUFA content in human milk was related to an outcome in the domain of neurodevelopment, body composition, or allergy/eczema. Observational studies were not excluded if the study also had an intervention. All studies included were accessed on 23 April 2021. Abstract screening was conducted independently by two authors and if there was no consensus, a third author screened and decided on eligibility for inclusion (Fig. 1).

The National Institutes of Health (NIH) Quality Assessment Tool for Observational Cohort and Cross-Sectional Studies[14] was used to score the certainty of evidence in studies. Criterion #7 from the NIH tool regarding timeframe sufficiency was not included in the validity assessment because this consideration was intrinsic to the systematic review design. The significance of association between PUFA predictors and health outcomes, the effect sizes for group differences in case-control studies, and considerations for covariates were weighted in assessing the certainty of evidence criteria. For each of the other 13 criteria, a score of + 1 was given if present in the study, 0 if it could not be reliably determined, or −1 if absent from the study. Scores were summed for individual studies and averaged across all studies within the neurodevelopmental, body composition, and allergy outcome domains. Certainty of evidence was graded for *n*-3 and *n*-6 in combination as high, moderate, low, or very low for average scores > 8.5, 8.5–4.5, 4.5–0.5, and < 0.5, respectively in each health outcome domain. The certainty of evidence was not scored for outcomes associated with the ratio of *n*-3/*n*-6 reported in studies; instead, this exposure was only assessed for considerations of inconsistency and imprecision. Inconsistency, indirectness, imprecision, and bias were assessed using the Grading of Recommendations Assessment, Development and Evaluation (GRADE) approach.[15] The odds ratio with confidence interval, mean of median difference, and r or r^2 value were reported when available.

Results

Studies retrieved and included in this systematic review were summarized (Tables 1–3).

Neurodevelopmental outcomes

N-3 fatty acids

In a study with mother-infant pairs (n = 39), human milk was obtained at one, three, and six months postpartum.[16] ALA, DHA, and total n-3 long chain PUFA content at one and three months were positively correlated with infant motor development at 6.6 months of age using the Child Development Scale. The correlation was still statistically significant after adjusting for infant age, sex, birthweight, maternal age, education, psychological status, and parity.[16] In another study, the Brazelton Neonatal Behavioral Assessment Scale (NBAS) was administered[17] at nine days of age to assess motor development. A positive correlation ($r = 0.57$) was found between relative DHA content in human milk and range of cluster score on the NBAS.[17] This finding suggests that infants receiving higher levels of DHA are less likely to frequently change state of arousal.

In a controlled intervention study, women were randomized to 2.2 g of DHA ($n = 98$) from week 20 gestation until birth or 4.0 g of olive oil ($n = 46$) daily[18] and human milk ($n = 78$) was collected at three days and six months postpartum. A positive correlation was found between relative DHA content in milk at three days postpartum for six out of seven subscales of Griffiths Mental Development Scales at 2.5 years of age: locomotor ($r = 0.27$), speech and hearing ($r = 0.288$), eye and hand coordination ($r = 0.41$), performance ($r = 0.32$), practical reasoning, and general quotient.[18] There were no correlations between health outcomes and DHA concentration in milk in the 6 month postpartum samples.[18]

Mother-infant pairs ($n = 709$) participated in a study aimed to identify pre- and postnatal determinants on child development and health outcomes. At two years of age, motor development assessed using the French Psychomotor Developmental Scale for Early Childhood of Brunet-Lézine and MacArthur Communicative Development Inventory (CDI) was not related to milk DHA content. At three years of age, there was no association observed between *n*-3 fatty acid content in milk and cognitive function assessed using the second French edition of

https://doi.org/10.1017/S2040174423000454 Published online by Cambridge University Press

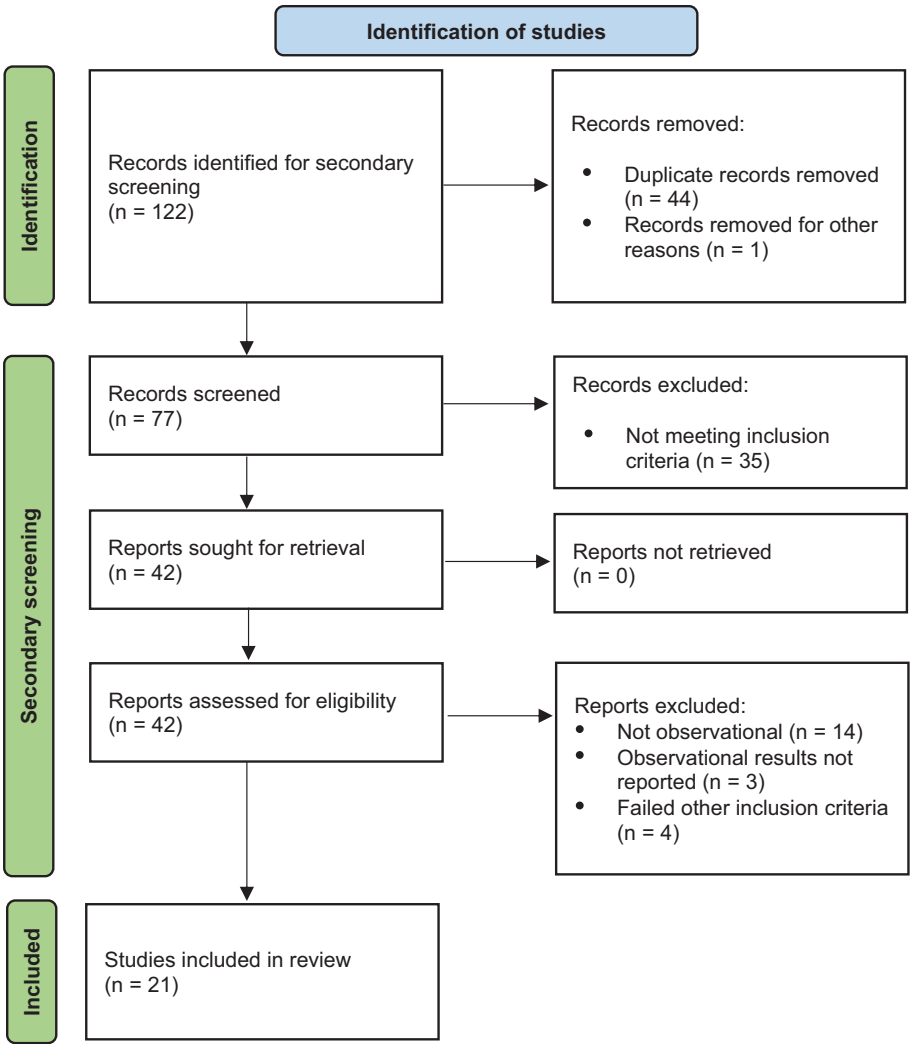

Figure 1. Flowchart of article identification, screening, and selection process for the systematic review.

the Ages and Stages Questionnaire (ASQ-3).[19] Furthermore, milk DHA content at four months was not related to cognitive development assessed by the Mullen Scales of Early Learning at 12 months,[20] and *n*-3 fatty acid content of colostrum and milk at one, two, and four months of lactation was not related to visual evoked potential at 2.5 or 7.5 months, or outcomes from the Bayley Scales of Infant Development (BSID)-II at 12 months.[21]

N-6 fatty acids

The relation between neurodevelopmental outcomes and *n*-6 fatty acid content was investigated in a study of human milk ($n = 78$) obtained at three days, six weeks, and six months postpartum. Infants were tested by a psychologist at 2.5 years for receptive language skills using the Peabody Picture Vocabulary Test IIIA.[18] Increased relative AA content in milk six months postpartum was associated with reduction in vocabulary skills including the average length of phrases used ($r = -0.533$) and number of words used ($r = -0.371$).[18] Relative LA content in milk one-week postpartum was negatively correlated to Motor-2 at two years assessed using the CDI and the ASQ-3 at three years of age.[19] Furthermore, milk AA content at four months was not related to cognitive development assessed by the Mullen Scales of Early Learning at 12 months,[20] and *n*-6 fatty acid content of colostrum and milk at one, two, and four months of lactation was not related to visual evoked potential at 2.5 or 7.5 months, or outcomes from the BSID-II at 12 months.[21]

N-3/n-6 fatty acid ratio

Fatty acids were measured in colostrum ($n = 319$) obtained 48–96 hours postpartum and correlated with the BSID-I at 14 months of age. The ALA/LA, EPA/AA, and DHA/AA ratios were positively correlated to mental development using the BSID-I at 14 months of age.[22] In a cohort study of infant-mother pairs ($n = 709$), cognitive function was assessed using the second French edition of the ASQ-3 relative to milk fatty acid content obtained one-week postpartum. Consistent with studies relating *n*-3 and *n*-6 fatty acid individually to neurodevelopment outcomes, a positive association between relative total *n*-3/*n*-6 ratio and ASQ-3 score at three years was observed.[19] Mothers with rs2397142 CC genotype in *ELOVL5* conferring higher DHA/AA ratio showed improved child cognitive index assessed by the McCarthy Scales of Children's Abilities at 14 months compared to G allele carriers.[23]

https://doi.org/10.1017/S2040174423000454 Published online by Cambridge University Press

Table 1. Studies of neurodevelopment outcomes

Study	Population	Sample Size	Assessment	Group Comparisons	Results
Zielinska et al. (2019)[16]	Women > 19 y with child > 6 wk in central urban Poland planning exclusive human milk feeding.	39 mother-infant pairs.	Children Development Scale measured at 6.6 months +/ −0.2 months.	Children exclusively breastfed for 6 mo. Milk sampled at 6 mo.	Positive correlation between DHA, ALA, *n*-3 PUFA, and motor development. Positive correlation between DHA and Perception subscale.
Hart et al. (2006)[17]	Mothers of healthy, full-term babies recruited from the maternity unit of university hospital, shortly after giving birth.	20 mother-infant pairs.	Brazelton Neonatal Behavioral Assessment scale at 9 d.	Milk obtained at 9 d and all participants were exclusively breast fed.	Positive relation between DHA and Range of State Cluster Score.
Dunstan et al. (2007)[18]	Atopic women from Western Australia between 16 and 20 weeks gestation with allergic rhinitis and/ or asthma, one or more positive skin prick tests to common allergens.	98 mother-infant pairs.	Asthma, atopic dermatitis, food allergy, sensitization measured at 12 mo. Griffiths mental development scales, Peabody Picture Vocabulary Test IIIA, and Child Behavior Checklist at 2.5 y.	Mixed feedings and infants exclusively breastfed for at least 6 mo.	EPA & DHA positively correlated to Griffiths scales at 2.5 y. AA negatively correlated to vocabulary (phrase length & number words used) at 6 mo.
Bernard et al. (2015)[19]	French EDEN mother-child cohort study. Participants were pregnant with amenorrhea before 24 wk gestation. Recruitment began in 2003 in Poitiers and Nancy university hospitals, and continued for 27 months.	709 children.	French Psychomotor Developmental Scale for Early Childhood of Brunet-Lezine & MacArthur Communicative Development Inventory at 2 y, Ages and Stages Questionnaire (ASQ-3) at 3 y.	Stratified by never breastfed, breastfed with low LA, breastfed with high LA content in milk.	Linoleic acid and total *n*-6 in colostrum was negatively related to Motor-2 and ASQ-3. Children in low LA level group scored higher on Motor-2 and ASQ-3 than other 2 groups. No relation between AA or DHA and motor or cognition scores. Negative relation between total *n*-6/*n*-3 PUFA ratio and ASQ score.
Keim et al. (2012)[20]	Women who attended prenatal clinics<20 wk pregnant at UNC Hospitals 20042005.	358 pregnant women and their children.	Mullen Scales of Early Learning at 12 mo.	Breastfeeding exclusivity up to 4 mo.	AA and DHA concentrations in milk at 4 mo was not associated with neurodevelopment in infants.
Hurtado et al. (2015)[21]	Pregnant mothers in Spain 20092010.	110 mother-infant pairs.	Bayley Scales of Infant Development-II at 12 months; visual evoked potential at 2.5, 7.5 mo.	Milk obtained at delivery, and 1, 2, 4 mo.	No relation between PUFA and outcomes.
Guxens et al. (2011)[22]	Pregnant women recruited in third trimester.	Mothers & 504 children.	Bayley Scales of Infant Development at 14 mo.	Cumulative intensity of breastfeeding and length of exclusive breastfeeding.	Higher ALA/LA ratio, DHA/AA ratio, DHA and *n*-3/*n*-6 in colostrum positively associated with mental development.
Morales et al. (2011)[23]	Mother-infant pairs who participated in either INMA-Sabadell and INMA-Menorca.	740 infant-mother pairs.	Bayley Scales of Infant Development, 1st Edition at 14 mo; McCarthy Scales of Children's Abilities at 4 y.	Elongase and desaturase SNPs. Five stratified groups: never, very short term, short term, long term breastfeeding, very long term breastfed.	Mothers with allele for higher EPA/AA ratio had child with higher cognition scores. Higher milk EPA/AA, DHA/AA related to higher cognition scores at 14 mo.

Body composition

N-3 fatty acids

Milk samples were obtained from mothers ($n = 208$) at six weeks and four months postpartum in the Impact of Nutritional Fatty acids during pregnancy and lactation for early human Adipose Tissue development (INFAT) study.[24] There was a positive correlation between relative EPA content ($r = 0.20$), DHA content, and total *n*-3 long chain PUFA in the six week milk with skinfold thickness (SFT) at 12 months ($r = 0.16$). Total *n*-3 long chain PUFA was also positively correlated to the ratio of subcutaneous to preperitoneal fat ($r = 0.12$) at six weeks of age, and with ponderal index (PI) at 12 months of age.[24] At one year postpartum, PI and body mass index (BMI) were related to DHA content ($r = 0.15$), but only PI was related to EPA content.[24] A follow-up to this

https://doi.org/10.1017/S2040174423000454 Published online by Cambridge University Press

Table 2. Studies of body composition outcomes

Study	Population	Sample size	Assessment	Group comparisons	Results
Much et al. (2013)[23]	Pregnant, healthy women in 15th week of gestation were recruited.	Milk available from 152 women at 6 wk and 120 women at 4 mo. Milk fatty acid profiles available for 56 infants at 4 mo and 31 infants at 12 months pp.	Sum of skinfold thickness (SFT) at 12 mo. At wk, 4 mo, 12 mo: fat mass, body fat, ratio, subcutaneous fat to preperitoneal fat, weight, length, BMI, ponderal index, lean body mass.	Regression adjusted for breastfeeding status at 6 wk, 4 mo, and 1 y.	DHA, EPA, LCPUFA *n*-3 in 6 wk milk positively related to SFT at 1 yr. AA and *n*-6 LCPUFA in 6 wk milk negatively related to BMI, lean body mass, weight until 4 mo.
Meyer et al. (2019)[25]	Women with a pre-pregnancy BMI 1830 recruited before 15 wk gestation.	169 mother-infant pairs.	Body weight, BMI, SFT, lean body mass at 2, 3, 4, 5 y.	Analyses adjusted for milk feeding (partially, or exclusively breastfed for 4 mo).	Positive relation between *n*-3 LCPUFA and lean body mass through 5 y. Positive relation between *n*-3/*n*-6 ratio with weight and BMI at 2 y, and lean body mass at 4 and 5 y.
Prentice et al. (2015)[26]	Mothers in early pregnancy were recruited from an antenatal center located in Cambridge, UK.	614 mothers provided milk samples when infants were 48 wk.	Weight, length, SFT at birth, 3, 12 mo.	Considered feeding type at 8 weeks (exclusive, mixed).	Linoleic acid inversely related to infant adiposity at 12 mo.
Ellsworth et al. (2020)[27]	Mothers who were either overweight/ obese or of normal weight pre-pregnancy in Ann Arbor, Michigan, USA.	55 mother-infant pairs.	Anthropometrics at 0.5, 2 and 6 mo.	Adjusted for feeding type (exclusive milk, mixed feeding).	*N*-6/*n*-3 LCPUFA ratio positively associated with weight-for-age *z*-score, weight-for-length *z*-score, BMI change from 0.56 mo across groups, but not in subgroup analysis of exclusive human milk feeding group.

study[24] was conducted in children ($n = 169$) who participated in the original study. Relative total *n*-3 from milk six-week postpartum was positively correlated to weight, BMI, SFT, and body fat at two years of age. Only the association between total *n*-3 fatty acids and lean body mass (LBM) remained statistically significant until five years of age. In the milk four months postpartum, total *n*-3 long chain PUFA concentration was negatively correlated to preperitoneal fat mass at two but not at three, four, or five years of age.[25]

N-6 fatty acids

Body composition outcomes were related to *n*-6 fatty acids in the INFAT study.[24] There were negative correlations between total relative *n*-6 long chain PUFA concentration in milk six weeks postpartum and BMI, LBM, PI, and weight at four months of age ($r = -0.41, -0.36, -0.31, -0.36$ respectively). *N*-6 long chain PUFA content was negatively correlated to fat mass at six weeks of age ($r = -0.29$), sum of SFT at 12 months of age ($r = -0.19$), and body fat percentage at six weeks of age ($r = -0.18$) but not at 12 months of age.[24] A negative correlation was observed between relative AA content in milk at six weeks postpartum and LBM, BMI, PI, and weight at four months of age ($r = -0.33, -0.35, -0.26, -0.32$ respectively). AA content of milk was negatively correlated to fat mass at six weeks of age ($r = -0.23$), but not at 12 months.[24] In another study, mothers ($n = 614$) provided a two week milk expression between four and eight months postpartum until 100 mL was collected. Absolute linoleic acid content was negatively related to SFT at 12 months of age, suggesting that n-6 fatty acid content of milk is negatively related to indices for body fat.[26]

N-3/n-6 fatty acid ratio

Body weight, BMI, SFT, and LBM were measured in children ($n = 169$) at two, three, four, and five years of age.[25] The *n*-3/*n*-6 long chain PUFA ratio from milk six weeks postpartum was positively associated with weight and BMI at two years of age, and negatively with LBM at four and five years of age.[25] In another study, the fatty acid content of milk at two weeks postpartum from mothers with BMI ≥ 25 ($n = 56$) was assessed as a predictor of child body composition and anthropometry at two weeks, two months and six months of age. In children receiving formula and human milk, the *n*-3/*n*-6 ratio was positively correlated with weight-for-age z-score, weight-for-length z-score, and BMI z-score change from two weeks to six months.[27] The clinical relevance of this finding is obscured because the *n*-3/*n*-6 ratio was not related to anthropometry in babies fed human milk exclusively.[27]

Allergy, skin & atopy outcomes

N-3 fatty acids

A study of human milk ($n = 315$) at one month postpartum assessed eczema, atopic dermatitis, and allergic sensitization at ages seven, 12, and 24 months. Higher total *n*-3 long chain PUFA concentrations were associated with lower risk for atopic dermatitis (OR = 0.24, 0.07–0.91) at two years of age and risk of allergic sensitization at one year of age (OR = 0.17, 0.04–0.77).[28] Another study examined the relation between incidence of atopic disease and relative *n*-3 concentration in human milk ($n = 263$)

https://doi.org/10.1017/S2040174423000454 Published online by Cambridge University Press

Table 3. Studies of allergy, skin, and atopy outcomes

Study	Population	Sample size	Assessments	Group comparisons	Results
Thijs et al. (2011)[28]	Healthy, pregnant women that participated in the KOALA birth cohort study & developed pregnancy-related pelvic girdle pain.	315 mother-infant pairs.	Eczema questionnaire at 6, 12, and 24 mo. IgE allergic sensitization at 12 and 24 mo.	Duration/exclusively of feeding, fatty acid supplement, season of collection included in analyses of milk from 1 mo. Adjusted for whether participants were in the conventional or alternative recruitment group.	Greater n-3 LCPUFA concentration in human milk correlated with decreased risk parent-reported eczema and atopic dermatitis at 2 y. Also correlated to decreased risk of allergic sensitization at 1 y.
Oddy et al. (2006)[29]	Infants who have a family history of allergy (one or both parents have asthma, hay fever, or eczema).	91 atopic mothers.	Skin prick test at 6 mo and 5 y. Skin condition reported at 1.5, 6, 12 months, and then annually until 5 y.	Obtained milk at 6 wk and 6 mo and characterized exclusivity of breastfeeding at 1 wk and confirmed some human milk consumption in all babies still at 6 mo.	Higher *n*-6 and *n*-6/*n*-3 ratio associated with greater risk for non-atopic eczema at 6 wk and 6 mo. No association between maternal asthma, infant asthma, and milk fatty acids.
Warstedt et al. (2016)[30]	Pregnant women who were at risk of having an allergic infant, living in Southern Sweden.	95 pregnant women and their infants.	Clinical exam at 3, 6, 12 mo. Skin prick tests at 6, 12, 24 mo for IgE-associated disease.	Milk at 3 d, 6 wk, 6 mo. Breastfeeding only; divided into quartiles of omega-3 proportions in milk; subgrouped by exclusive breastfeeding at 3 mo vs. non-exclusive.	Lower IgE among children related to higher EPA and DHA, and lower AA/EPA ratio in colostrum. Lower IgE among children related to higher EPA and DHA at 1 mo.
Hoppu et al. (2005)[31]	Pregnant women with atopic disease enrolled at 35-36 w of gestation.	34 mother-infant pairs.	Clinical examination and atopic disease diagnosis at 1, 3, 6, 12 mo.	Milk obtain at 1 month and exclusivity and duration of breastfeeding reported. Duration was similar for infants with and without atopy.	Total *n*-3 PUFA and EPA lower in infants that developed atopic disease.
Rosenlund et al. (2016)[32]	Families in Sweden from 2004 to 2007.	330 children. 245 mothers gave milk samples.	Blood tests for allergen-specific IgE levels performed at 6, 12, 24 mo.	Milk obtained at 2 mo and analyses adjusted for whether the infant was exclusively breastfed at 2 mo.	Inverse relation between milk *n*-3 PUFA content and sensitization. Higher AA/EPA ratio associated with higher risk of sensitization.
Wijga et al. (2006)[33]	Allergic mothers (displayed asthma, hay fever, and/or other allergy) or nonallergic (no symptoms).	265 mother-infant pairs.	Questions adapted from international study on asthma and allergy in children at 1 and 4 y.	Time milk sample was collected (average ~ 1516 wk).	Higher n-3 PUFA and *n*-3/*n*-6 PUFA ratio associated with low eczema, asthma and persistent symptoms. Inverse relation between AA and allergic symptoms for children of mothers with allergy.
Hua et al. (2018)[34]	Pregnant women with and without atopic diseases and/or allergies.	443 mother and 445 infants.	Questionnaire and clinical exam at 2, 4, 6, 12, 18, 24 mo for atopic disease.	Obtained colostrum and milk at 2 mo and reported duration of exclusive breastfeeding.	No correlation between *n*-6, *n*-3, or n-6/n-3 ratio and atopic disease.
Morales et al. (2012)[41]	Pregnant women in third trimester at enrollment.	580 infants.	Physician diagnosis of atopic eczema at 6, 14 mo.	Five stratified groups: never, very short term, short term, long term breastfeeding, very long term breastfed.	No associations between fatty acids and atopic eczema.

collected at six weeks and six months postpartum.[29] Infants whom developed non-atopic eczema at six months of age had lower total *n*-3 PUFA in the milk six weeks postpartum, but not in the milk six months postpartum.[29]

Women at risk of having a child with allergy were randomized to receive 1.6 g of EPA and 1.1 g DHA or soya bean placebo from 25 weeks of gestation to three months postpartum ($n = 145$).[30] Milk samples were collected at two-four days, one month, and three months postpartum. At two years of age, IgE-associated disease was lower in children consuming higher levels of EPA and DHA in milk from two-four days and one month postpartum.[30] In another study of mother-infant pairs ($n = 34$), the relation between atopic disease and fatty acid composition of milk obtained at one month postpartum was investigated.[31] Lower content of EPA and total *n*-3 PUFA was observed in milk from mothers with infants who developed atopic dermatitis within the first year of life.[31] In another cohort ($n = 245$), the risk for developing allergic sensitization up to 24 months of age was lower in children consuming milk with higher total n-3 content.[32]

N-6 fatty acids

Human milk ($n = 265$) was obtained from mothers with or without asthma, hay fever, allergy for pets, or allergy for house dust/dust mites at three months postpartum.[33] The International Study on Asthma and Allergy in Children questionnaire was completed by parents at one and four years of child age. Indicators of allergic disease were determined by eczema at one year, and asthma and sensitization at four years. Lower AA content in milk was a risk factor for all allergic symptoms at one and four years of age only in children of mothers with allergy.[33] In addition, the total *n*-6 relative concentrations in milk six weeks postpartum were higher in a study of infants ($n = 263$) whom developed non-atopic eczema at six months of age.[29]

N-3/n-6 fatty acid ratio

Infants with at least one parent with a history of allergy defined by hay fever, eczema, and/or asthma had skin conditions reported at six weeks, six months, 12 months, and annually until five years of age.[29] Decreased *n*-3/*n*-6 ratio in milk six weeks postpartum was associated with increased risk of non-atopic eczema at six weeks and six months but not at one or five years of age.[29] High *n*-3/*n*-6 ratio was associated with lower incidence of eczema at one year, asthma at four years, and persistence of symptoms assessed by a questionnaire based on the International Study on Asthma and Allergy in Children.[33] Blood samples were collected from infants ($n = 245$) at six, 12, and 24 months of age and infants were assessed for sensitization to any of hen's egg, cow's milk, peanut, cat, dog, birch and timothy, determined as IgE $\geq$ 0.35 kUA/l. Increased risk of sensitization up to 24 months was associated with decreased EPA/AA ratio from milk two months postpartum.[32] Lower incidence of IgE-associated disease was observed in infants at six, 12, and 24 months of age with higher EPA/AA in colostrum, but not with fatty acids in milk from one and three months postpartum.[30]

Another study indicated no association between *n*-3/*n*-6 ratio in milk with atopic dermatitis.[34] Colostrum and milk two months postpartum were obtained from mothers ($n = 146$) with a history of allergy. Infants were assessed at two, four, six, eight, 12, and 24 months of age for atopic dermatitis. There were no statistically significant findings observed for fatty acid concentration in milk and infant atopic dermatitis when compared to healthy control groups with and without controlling for potential confounders including maternal age and allergy, cesarean section, parity, pets in the household, tobacco smoking within the last year, and use of antibiotics/probiotics during sample collection.[34]

Certainty of evidence

Quality assessment

Risk of bias was present in studies where analyses were conducted *post hoc* or with convenience sampling. Furthermore, bias was present when a 24 hr expression of milk was not collected or reported. Inconsistency and imprecision were considered negligible as the fatty acid predictors were directionally and uniformly associated with the health outcomes in the three domains of interest in 20 of 21 studies assessed. Indirectness was present for each health outcome assessed. For example, neurodevelopmental outcome domains included visual and cognitive indices across a range of data acquisition tools. Body composition was assessed by various metrics including weight, PI, or BMI. Finally, allergic outcomes included serum IgE antibody titers, and skin problems, atopy, eczema, and asthma which were often self-reported.

The average certainty of evidence scores for neurodevelopment, body composition, and allergy, skin & eczema were 8.0, 8.5, and 7.6 respectively. There was evidence of *moderate* certainty from the systematic review of observational studies demonstrating an association between *n*-3 and *n*-6 fatty acids and neurodevelopmental outcomes. Assessment of the observational studies on body composition outcomes yielded evidence of *moderate* certainty from the systematic review for the association to *n*-3 and *n*-6 fatty acids. Imprecision and indirectness were evident mostly in studies of allergic outcomes. There was evidence of *moderate* certainty from the systematic review of observational studies demonstrating an association between *n*-3 and *n*-6 and allergy outcomes.

Discussion

Increased total *n*-3, DHA, EPA, and ALA were associated positively with developmental health outcomes and growth and adiposity indices. *N*-3 fatty acids were associated with lower risk of allergy, asthma and eczema. Increased total *n*-6, AA, and LA were negatively associated with developmental health outcomes and growth and adiposity indices. *N*-6 fatty acids were associated with higher risk for issues with skin and allergic manifestation. These findings were corroborated by studies that analyzed how the ratio of *n*-3/*n*-6 fatty acid correlated with neurodevelopmental, body composition, and allergy outcomes.

The certainty of evidence assessment in this systematic review was also corroborated by evidence from clinical trials. Formula containing dietary AA at 0.32% of fatty acids supported visual acuity assessed at 12 months of age when DHA was also present in an equal but not greater amount.[35] In addition, intake of AA at higher levels may lower the number of B cells and B cell activation markers in children at 13 weeks[2] impacting immune functions. Mothers supplemented with 6 g of fish oil had infants with a higher BMI z-score and PI at three months compared to those of mothers given olive oil.[36] A systematic review and meta-analysis showed that postnatal supplementation of mothers with fish oil resulted in increases in offspring birth weight and waist circumference.[37] Newborn growth and development are supported by specific levels of PUFA intake in mothers in the perinatal period. Understanding optimal intake levels of *n*-3 and *n*-6 fatty acids in children is required to continue to support health outcomes throughout early life.

https://doi.org/10.1017/S2040174423000454 Published online by Cambridge University Press

Other child health outcomes related to PUFA content in milk

Observational studies investigating the impact of early nutrition on respiratory outcomes is needed. There is a link between breastfeeding and decreased risk of respiratory illness later in life[38] which may be due to *n*-3 and *n*-6 fatty acid content of milk. A narrative review concluded that *n*-3 long chain PUFA supplementation lowers the risk of respiratory illness.[39] Another study found that a DHA supplement resulted in significantly lower prevalence of asthma and persistent wheezing in children aged three to five years.[40] Similarly, observational research assessing the relation of early life nutrition to gastrointestinal outcomes is limited. A study assessed how the relative DHA content of human milk ($n = 580$) obtained 48-96 hr postpartum was related to the incidence of gastroenteritis by parent-reported questionnaire.[41] Increased DHA content was associated with reduced risk of gastroenteritis at six months of age (OR = 0.25, 0.11–0.57) and for recurrent gastroenteritis (OR = 0.28, 0.09–0.81).[41] A review of studies relating *n*-3 and *n*-6 fatty acid content in milk to respiratory and gastrointestinal outcomes in early life is warranted in future.

Timeframe

The tool used to assess certainty of evidence[14] in this systematic review suggests that a determination should be made regarding the appropriateness of the timeframe for which an outcome is associated with the exposure. The authors did not assess the studies individually to determine whether the timeframe of 0–5 years was sufficient to reasonably expect to see an association between PUFA content in milk and the three health domains of interest. Instead, the assessment was made holistically and incorporated into the study design to inform the inclusion criteria. It is reasonable to conclude that there is a link between early nutrition with neurodevelopment, body composition, and allergy outcomes in the first 5 years of life. This is evidenced in a study of infants who were exclusively breastfed for 16 weeks whom were observed to have lower incidence of atopic dermatitis than infants whom did not consume human milk exclusively.[42] In another study, children who were exclusively breastfed in infancy had significantly higher cognition, verbal intelligence, and preschool language scores at four years than children consuming formula.[43] Early life nutrition impacts short and medium-term adiposity, a facet of body composition, and there is a link between infant nutrition and body composition in individuals up to 8 years of age.[44] Thus, it is reasonable to conclude that there is a link between fatty acid nutrition in infancy and early childhood to neurodevelopmental, body composition, and allergic outcomes.

Maternal supplementation with DHA during pregnancy is associated with higher cord blood DHA and presumably with supply of DHA to the fetus.[45] However, supplementation of mothers from 21 weeks of gestation until birth with 800 mg of DHA/day did not improve cognitive and language composite scores of children at 18 months compared to the placebo group.[46] Furthermore, in a study of maternal DHA supplementation (400 mg/day) from 20 weeks of lactation until six months postpartum, child neurodevelopment at 12 months was not different from the placebo group.[47] A recent review suggests that supply of DHA to newborns has stronger effects compared to maternal supplementation in pregnancy.[48] The 2020 US Dietary Guidelines Advisory Committee Report also reviewed evidence comprehensively to conclude that antenatal *n*-3 PUFA supplementation showed little benefit on growth and neurodevelopment outcomes.[49] Intervention with *n*-3 fatty acids is considered more effective when targeted to newborn. Developing a comprehensive understanding of the maternal factors that affect synthesis and biodistribution of long chain PUFA to milk will be critical to informing interventions promoting healthy growth and development in newborns.

Absolute and relative concentrations of fatty acids

All but one study in this systematic review utilized relative fatty acid concentrations in the analysis of health outcomes. The use of absolute versus relative concentrations of fatty acids in studies is not believed to have a significant impact on the results.[50] In one study, there was no significant relation between all-cause mortality and plasma phospholipid fatty acid content when analyzed in absolute or relative amounts. For this systematic review, the sparsity of reporting or analyzing the absolute concentration of fatty acids was believed not to impact the certainty of evidence assessment. However, presenting data in absolute fatty acid concentrations may help to prevent loss in publication of datasets for secondary analyses.[50]

Limitations in the studies and systemic review

An extensive review on this topic that distinctly notes fatty acid content (relative vs absolute amount), control for covariates (age, sex, etc.), and genotype (FADS, ELOVL) has not yet been published. Study populations were small in some cases and outcomes were self-reported by parents which allows potential bias. This systematic review did not differentiate between colostrum, transitional, and mature milk. In addition, essential fatty acids ALA and LA were not distinguished from long chain products EPA, DHA, and AA for the certainty of evidence assessment. In many of the cohort studies, milk was collected at multiple time points or outcomes were assessed at multiple times. A predictor that was statistically significantly related to an outcome in only a single instance in a study was sufficient to ascribe an association for the purposes of the systematic review.

Overall, the studies assessed in this review primarily measured health outcomes when children were toddler- (one to three years) or preschooler-aged (three to five years). Over half of the studies measured health outcomes when children were one year (12 months) of age. In addition, nearly half of the studies assessed health outcomes in infants between ages two to three years and at six months. Approximately one-quarter of studies observed health outcomes in infants at 2.5 months or between three and four months. Few studies measured health outcomes at 1–2 years or from four to five years of age.

Conclusion

N-3 fatty acids in human milk were consistently positively related to neurodevelopment, growth & body composition, and protection from allergic, skin & atopy outcomes in young children. *N*-6 fatty acids in milk were consistently related with negative attributes for health outcomes in the three domains of interest. The certainty of evidence was *moderate* for the relations between *n*-3 and *n*-6 fatty acid content of milk with neurodevelopment, body composition, and allergy. Longer term studies investigating dose-response relations and whether health outcomes associated with human milk PUFA content persist beyond childhood are warranted.

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/S2040174423000454.

https://doi.org/10.1017/S2040174423000454 Published online by Cambridge University Press

Acknowledgments. None.

Financial support. None.

Competing interests. None.

Ethical standards. Omitted due to systematic review.

References

1. Innis SM. Impact of maternal diet on human milk composition and neurological development of infants123. *Am J Clin Nutr*. 2014; 99(3), 734S–741S.
2. Miklavcic JJ, Larsen BMK, Mazurak VC, *et al.* Reduction of arachidonate is associated with increase in b-cell activation markers in infants: a randomized trial. *J Pediatr Gastroenterol Nutr*. 2017; 64(3), 446–453.
3. Makrides M, Neumann MA, Gibson RA. Effect of maternal docosahexaenoic acid (DHA) supplementation on breast milk composition. *Eur J Clin Nutr*. 1996; 50(6), 352–357.
4. Ueno HM, Higurashi S, Shimomura Y, *et al.* Association of DHA concentration in human breast milk with maternal diet and use of supplements: a cross-sectional analysis of data from the Japanese human milk study cohort. *Curr Dev Nutr*. 2020; 4(7), nzaa105.
5. Chamorro R, Bascunan KA, Barrera C, Sandoval J, Puigrredon C, Valenzuela R. Reduced n-3 and n-6 PUFA (DHA and AA) concentrations in breast milk and erythrocytes phospholipids during pregnancy and lactation in women with obesity. *Int J Environ Res Public Health*. 2022; 19(4), 1930.
6. Videla LA, Hernandez-Rodas MC, Metherel AH, Valenzuela R. Influence of the nutritional status and oxidative stress in the desaturation and elongation of n-3 and n-6 polyunsaturated fatty acids: impact on the non-alcoholic fatty liver disease. *Prostaglands Leukot Essent*. 2022; 181, 102441.
7. Koletzko B, Bergmann K, Brenna JT, *et al.* Should formula for infants provide arachidonic acid along with DHA? A position paper of the european academy of paediatrics and the child health foundation. *Am J Clin Nutr*. 2020; 111(1), 10–16.
8. Berman DR, Liu YQ, Mozurkewich E. Docosahexaenoic acid confers neuroprotection in a rat model of perinatal hypoxia-ischemia potentiated by Escherichia coli lipopolysaccharide-induced systematic inflammation. *Am J Obstet Gynecol*. 2010; 202(5), 469.e1–469.e6.
9. Clandinin MT, Chappell JE, Leong S, Heim T, Swyer PR, Chance GW. Intrauterine fatty acid accretion rates in human brain: implications for fatty acid requirements. *Early Hum Dev*. 1980; 4(2), 191–129.
10. Reyes SM, Brockway M, McDermid JM,. Human milk micronutrients and child growth and body composition in the First 2 years: a systematic review. *Adv Nutr*. 2023, 100082. doi: 10.1016/j.advnut.2023.06.005
11. National Academies of Sciences, Engineering, and Medicine. *Scanning for New Evidence on the Nutrient Content of Human Milk: A Process Model for Determining Age-Specific Nutrient Requirements*, 2020. The National Academies Press, Washington, DC.
12. Miklavcic J, Mitguard S, Doucette O. *Fatty Acids & Early Life Outcomes*, 2023. PROSPERO, York, UK. https://www.crd.york.ac.uk/prospero/display_record.php?ID=CRD42023400059.
13. Page MJ, McKenzie JE, Bossuyt PM, *et al.* The prisma 2020 statement: an updated guideline for reporting systematic reviews. *BMJ*. 2021; 372(71).
14. National Heart, Lung, and Blood Institute. *Study Quality Assessment Tools*, 2021. National Institutes of Health, Bethesda, MD. https://www.nhlbi.nih.gov/health-topics/study-quality-assessment-tools.
15. Granholm A, Alhazzani W, Moller MH. Use of the GRADE approach in systematic reviews and guidelines. *Br J Anaesth*. 2019; 123(5), 554–559.
16. Zielinska M, Hamulka J, Grabowicz-Chądrzyńska I, Brys J, Wesolowska A. Association between breastmilk LC PUFA, carotenoids and psychomotor development of exclusively breastfed infants. *Int J Environ Res Public Health*. 2019; 16(7), 1144.
17. Hart SL, Boylan LM, Carroll SR, *et al.* Brief report: newborn behavior differs with docosahexaenoic acid levels in breast milk. *J Pediatr Psychol*. 2006; 31(2), 221–226.
18. Dunstan JA, Mitoulas LR, Dixon G, *et al.* The effects of fish oil supplementation in pregnancy on breast milk fatty acid composition over the course of lactation: a randomized controlled trial. *Pediatr Res*. 2007; 62(6), 689–694.
19. Bernard JY, Armand M, Garcia C, *et al.* The association between linoleic acid levels in colostrum and child cognition at 2 and 3 y in the EDEN cohort. *Pediatr Res*. 2015; 77(6), 829–835.
20. Keim SA, Daniels JL, Siega-Riz AM, Herring AH, Dole N, Scheidt PC. Breastfeeding and long-chain polyunsaturated fatty acids intake in the first 4 post-natal months and infant cognitive development: an observational study. *Matern Child Nutr*. 2012; 8(4), 174–182.
21. Hurtado JA, Iznaola C, Pena M, *et al.* Effects of maternal omega-3 supplementation on fatty acids and on visual and cognitive development. *J Pediatr. Gastroenterol Nutr*. 2015; 61(4), 472–480.
22. Guxens M, Mendez MA, Molto-Puigmarti C, *et al.* Breastfeeding, long-chain polyunsaturated fatty acids in colostrum, and infant mental development. *Pediatrics*. 2011; 128(4), 880–889.
23. Morales E, Bustamante M, Gonzalez JR, *et al.* Genetic variants of the FADS gene cluster and ELOVL gene family, colostrums LC-PUFA levels, breastfeeding, and child cognition. *PLoS One*. 2011; 6(2), 6.
24. Much D, Brunner S, Volhardt C, *et al.* Breast milk fatty acid profile in relation to infant growth and body composition: results from the INFAT study. *Pediatr Res*. 2013; 74(2), 230–237.
25. Meyer DM, Brei C, Stecher L, Much D, Brunner S, Hauner H. Associations between long-chain PUFAs in maternal blood, cord blood, and breast milk and offspring body composition up to 5 years: follow-up from the INFAT study. *Eur J Clin Nutr*. 2019; 73(3), 458–464.
26. Prentice P, Ong KK, Schoemaker MH, *et al.* Breast milk nutrient content and infancy growth. *Acta Paediatr*. 2015; 105(6), 641–647.
27. Ellsworth L, Perng W, Harman E, Das A, Pennathur S, Gregg B. Impact of maternal overweight and obesity on milk composition and infant growth. *Matern Child Nutr*. 2020; 16(3), e12979.
28. Thijs C, Muller A, Rist L. Fatty acids in breast milk and development of atopic eczema and allergic sensitisation in infancy. *Allergy*. 2011; 66(1), 58–67.
29. Oddy WH, Pal S, Kusel MMH, *et al.* Atopy, eczema and breast milk fatty acids in a high-risk cohort of children followed from birth to 5 yr. *Pediatr Allergy Immunol*. 2006; 17(1), 4–10.
30. Warstedt K, Furuhjelm C, Falth-Magnusson K, Fageras M, Duchen K. High levels of omega-4 fatty acids in milk from omega-3 fatty acid-supplemented mothers are related to less immunoglobulin E-associated disease in infancy. *Acta Paediatr*. 2016; 105(11), 1337–1347.
31. Hoppu U, Rinne M, Lampi A, Isolauri E. Breast milk fatty acid composition is associated with development of atopic dermatitis in the infant. *J Pediatr Gastroenterol Nutr*. 2005; 41(3), 335–338.
32. Rosenlund H, Fagerstedt S, Alm J, Mie A. Breastmilk fatty acids in relation to sensitization – the ALADDIN birth cohort. *Eur J Allergy Clin*. 2016; 71(10), 1444–1452.
33. Wijga AH, Van Houwelingen AC, Kerkhof M, *et al.* Breast milk fatty acids and allergic disease in preschool children: the prevention and incidence of asthma and mite allergy birth cohort study. *J Allergy Clin Immunol*. 2006; 117(2), 440–447.
34. Hua M, Su H, Kuo M, *et al.* Association of maternal allergy with human milk soluble CD14 and fatty acids, and early childhood atopic dermatitis. *Pediatr Allergy Immunol*. 2018; 30(2), 204–213.
35. Birch EE, Carlson SE, Hoffman DR, *et al.* The DIAMOND (DHA intake and measurement of neural development) study: a double-masked, randomized controlled clinical trial of the maturation of infant visual acuity as a function of the dietary level of docosahexaenoic acid. *Am J Clin Nutr*. 2010; 91(4), 848–859.
36. Satokar Derraik VV, Harwood JGB, etal M. Fish oil supplementation during pregnancy and postpartum in mothers with overweight and obesity to improve body composition and metabolic health during infancy: a double-blind randomized controlled trial. *Am J Clin*. 2023; 117(5), 883–895.
37. Li G, Chen H, Zhang W, Tong Q, Yan Y. Effects of maternal omega-3 fatty acid supplementation during pregnancy/lactation on body composition of

the offspring: a systematic review and meta-analysis. *Clin Nutr*. 2018; 37(5), 1462–1473.
38. Gardner AS, Rahman IA, Lai CT, *et al.* Changes in fatty acid composition of human milk in response to cold-like symptoms in the lactating mother and infant. *Nutrients*. 2017; 9(9), 1034.
39. Miles EA, Childs CE, Calder PC. Long-chain polyunsaturated fatty acids (LCPUFAs) and the developing immune system: a narrative review. *Nutrients*. 2021; 13(1), 247.
40. Bisgaard H, Stokholm J, Chawes BL, *et al.* Fish oil-derived fatty acids in pregnancy and wheeze and asthma in offspring. *N Engl J Med*. 2016; 375(26), 2530–2539.
41. Morales E, Garcia-Esteban R, Guxens M, *et al.* Effects of prolonged breastfeeding and colostrum fatty acids on allergic manifestations and infections in infancy. *Clin Exp Allergy*. 2012; 42(6), 918–928.
42. Schoetzau A, Filipiak-Pittroff B, Franke K, *et al.* Effect of exclusive breastfeeding and early solid food avoidance on the incidence of atopic dermatitis in high-risk infants at 1 year of age. *Pediatr Allergy Immunol*. 2002; 13(4), 234–242.
43. Bellando J, McCorkle G, Spray B, *et al.* Developmental assessments during the first 5 years of life in infants fed breast milk, cow's milk formula, or soy formula. *Food Sci Nutr*. 2020; 8(7), 3469–3478.
44. Bar S, Milanaik R, Adesman A. Long-term neurodevelopmental benefits of breastfeeding. *Curr Opin Pediatr*. 2016; 28(4), 559–566.
45. Gázquez A, Giménez-Bañón MJ, Prieto-Sánchez MT, *et al.* Self-reported DHA supplementation during pregnancy and its association with obesity or gestational diabetes in relation to DHA concentration in cord and maternal plasma: results from NELA, a prospective mother-offspring cohort. *Nutrients*. 2021; 13(3), 843.
46. Makrides M, Gibson RA, McPhee AJ, *et al.* Effect of DHA supplementation during pregnancy on maternal depression and neurodevelopment of young children: a randomized controlled trial. *JAMA*. 2010; 304(15), 1675–1683.
47. Khandelwal S, Kondal D, Chaudhry M, *et al.* Effect of maternal docosahexaenoic acid (DHA) supplementation on offspring neurodevelopment at 12 Months in India: a randomized controlled trial. *Nutrients*. 2020; 12(10), 3041.
48. Comitini F, Peila C, Fanos V, Coscia A. The docosahexanoic acid: from the maternal-fetal dyad to early life toward metabolomics. *Front Pediatr*. 2020; 8, 538.
49. Dietary Guidelines Advisory Committee. *Scientific Report of the 2020 Dietary Guidelines Advisory Committee: Advisory Report to the Secretary of Agriculture and the Secretary of Health and Human Services*, 2020. U.S. Department of Agriculture, Agricultural Research Service, Washington, DC.
50. Miura K, Hughes MCB, Ungerer JPJ, Smith DD, Green AC. Absolute versus relative measures of plasma fatty acids and health outcomes: examples of phospholipid omega-3 and omega-6 fatty acids and all-cause mortality in women. *Eur J Nutr*. 2018; 57(2), 713–722.

https://doi.org/10.1017/S2040174423000454 Published online by Cambridge University Press