European Journal of Clinical Nutrition
https://doi.org/10.1038/s41430-019-0484-z

ARTICLE

Epidemiology

Polyunsaturated fatty acids in middle childhood and externalizing and internalizing behavior problems in adolescence

Sonia L. Robinson[1] · Henry Oliveros[2] · Mercedes Mora-Plazas[3] · Constanza Marín[2,3] · Betsy Lozoff[4,5] · Eduardo Villamor[1]

Received: 6 May 2018 / Revised: 25 June 2019 / Accepted: 22 July 2019

Abstract

Background/Objectives We sought to determine the associations of n-3 and n-6 polyunsaturated fatty acids (PUFA) in middle childhood with externalizing and internalizing behavior problems in adolescence.

Subjects/Methods Using gas-liquid chromatography, we quantified n-3 and n-6 PUFA in serum samples of 444 Colombian schoolchildren aged 5–12 years at the time of enrollment into a cohort study. After a median 6 years, adolescent externalizing and internalizing behavior problems were determined with the Youth Self Report (YSR) questionnaire. We estimated adjusted mean behavior problem score differences with 95% confidence intervals (CIs) between quartiles of each PUFA using multivariable linear regression. We also considered as exposures the Δ6-desaturase (D6D) and Δ5-desaturase (D5D) enzyme activity indices.

Results Docosahexaenoic acid (DHA) was positively associated with externalizing problems; every standard deviation (SD) of DHA concentration was associated with an adjusted one unit higher externalizing problem score (95% CI: 0.1, 1.9). The D5D enzyme activity index was inversely related to externalizing problem scores. Alpha-linolenic acid concentration was positively associated with internalizing problem scores, whereas adrenic acid was inversely related to this outcome.

Conclusions Serum PUFA in middle childhood were related to behavior problems in adolescence. Some of these associations might reflect the role of D5D enzyme activity.

Introduction

One in five adolescents worldwide has an externalizing or internalizing disorder [1]. These disorders lie at the extreme end of a continuum of behavior problems, which are associated with adult outcomes including low educational attainment [2], psychiatric disorders [3], and criminality [4].

Omega-3 (n-3) and omega-6 (n-6) polyunsaturated fatty acids (PUFA) are essential for adequate neurodevelopment. The precursors of each family, alpha-linolenic acid (18:3n-3, ALA) and linoleic acid (18:2n-6, LA), are considered essential as they cannot be synthesized in humans and must be obtained from diet. ALA and LA are metabolized into longer chain PUFA endogenously; however, this conversion is inefficient. Several longer chain PUFA accumulate in cell membranes of the fetal brain [5] where they affect protein function, synaptogenesis, and monoamine neurotransmitter metabolism [6–8]. In addition, long-chain n-3 and n-6 PUFA are precursors to anti- and pro-inflammatory eicosanoids, respectively, which may exert opposing effects on the development of neuropsychiatric disorders [9–11].

Epidemiologic research on the role of PUFA on behavior has focused primarily on maternal or infant docosahexaenoic acid (22:6n-3, DHA) and arachidonic acid (20:4n-6, AA) status in relation to behavior problems in early or

Supplementary information The online version of this article (https://doi.org/10.1038/s41430-019-0484-z) contains supplementary material, which is available to authorized users.

✉ Eduardo Villamor
villamor@umich.edu

[1] Department of Epidemiology, University of Michigan School of Public Health, Ann Arbor, MI, USA

[2] Universidad de la Sabana, Chia, Colombia

[3] Foundation for Research in Nutrition and Health, Bogota, Colombia

[4] Center for Human Growth and Development, University of Michigan, Ann Arbor, MI, USA

[5] Department of Pediatrics and Communicable Diseases, University of Michigan Medical School, Ann Arbor, MI, USA

Published online: 05 August 2019

middle childhood, but the results have been inconsistent [12–16]. In a small ($n = 20$) randomized trial of children aged 6–12 years with major depressive disorder (MDD), those supplemented with eicosapentaenoic acid (22:5n-3, EPA)+DHA for 16 weeks had a reduction in depressive symptoms compared with children who received a placebo composed mostly of LA [17]. During adolescence, supplementation with n-3 PUFA resulted in improved behavior in trials from the United Kingdom [18] and Mauritius [19]. Meta-analyses of randomized trials conducted in adults with depressive disorders showed that the benefits of long chain n-3 PUFA supplementation against depressive symptoms were greater when the supplements contained a higher percentage of EPA than DHA; this suggests that a potential effect might be through EPA [20, 21]. However, studies in adults may not be directly applicable to children. No study has examined the relations of PUFA status during middle childhood and adolescent behavior problems.

The aim of this study was to investigate the associations between serum PUFA concentrations at ages 5–12 years and behavior problems in adolescence among Colombian schoolchildren. We hypothesized that serum concentrations of the long-chain n-3 PUFA EPA and DHA in middle childhood would be inversely related to adolescent behavior problems, whereas n-6 PUFA would be positively associated with these outcomes.

Subjects and methods

Study design and population

We conducted this study in the context of the Bogotá School Children Cohort, a prospective investigation in Bogotá, Colombia, which has been detailed previously [22]. In brief, in February 2006, we recruited 3202 schoolchildren aged 5–12 years through random selection from primary public schools in Bogotá. Parents or primary caregivers gave written informed consent. Children gave written assent to participate. The study protocol was approved by the Ethics Committee of the National University of Colombia Medical School. The Institutional Review Board at the University of Michigan (UM) approved the use of data.

At the time of enrollment, we obtained information on sociodemographic characteristics of children and their families through a parental self-administered questionnaire. Household socioeconomic status was determined per classification by the local government. Household food insecurity was assessed with a validated Spanish language version of the USDA Household Food Security Survey module [23, 24]. Trained research assistants measured children's height and weight at a scheduled school visit. Height was measured without shoes to the nearest 1 mm with a wall-mounted portable Seca 202 stadiometer (Seca, Hanover, MD), and weight was measured in light clothing to the nearest 0.1 kg with Tanita H5301 electronic scales (Tanita, Arlington Heights, IL). Height and weight were also measured among mothers present at schools. At the end of these visits, investigators collected fasting blood samples through antecubital venipuncture. The samples were transported to the Colombian National Institute of Health, where they were cryostored before transportation to the United States for analyses.

The evaluation of behavior outcomes has been described in detail elsewhere [25]. In brief, between 2011 and 2015 we conducted a follow-up assessment on a random sample of 1139 cohort participants that included the ascertainment of adolescent behavior problems using the Spanish language version of the Youth Self Report (YSR) [26]. The YSR, a self-administered questionnaire designed for use in 11–18 year olds [27], consists of 112 statements of behaviors or feelings that the adolescents may experience. From responses to these questions, we calculated age- and sex-standardized continuous scores for eight behavior problems subscales (aggressive and rule-breaking behavior; anxious/depressed, withdrawn/depressed, somatic complaints; and attention, social, and thought problems) using software provided by the test developer [28]. We then computed scores for total externalizing and internalizing problems, which are comprised of aggressive and rule-breaking behavior and anxious/depressed, withdrawn/depressed, and somatic complaints, respectively. The YSR has been widely used in other Latin American settings [26, 29, 30].

Laboratory methods

We quantified serum fatty acids (FAs) in baseline samples of approximately 20% of participants at the UM Metabolomics and Obesity Research Center. We prepared FA methyl esters of total lipids with BF3-methanol [31]. Methyl esters were extracted from a thin-layer chromatography plate and solvents were dried and resuspended in hexane. Approximately 2 µl of sample was injected via an autosampler and analyzed on a gas-liquid chromatography machine using a 100 m SP-2560 column with optimum conditions for separation (Model 6890N, Agilent, Santa Clara, CA). Eluted peaks were analyzed using the Chemstation software (Agilent). The concentration of each FA was determined using a calibration curve with C17:0 methyl ester as the standard.

Data analysis

In the subset selected for quantification of FA, 444 children completed the YSR. This sample size provides adequate

statistical power to detect previously reported differences in the continuous distributions of behavior outcomes [25] by levels of dichotomous exposures with prevalences ≥25%. Primary outcomes of interest were the continuous total externalizing and internalizing problem scores. Secondary outcomes were the behavior problems subscale scores that comprise these composite scores. The primary exposures were the percentage of total weight concentration of the n-3 ALA, 20:5n-3, EPA, docosapentaenoic acid (22:5n-3, DPA) and DHA; and the n-6 LA, gamma-linolenic acid (18:3n-6, GLA), eicosadienoic acid (20:2n-6), dihomo-gamma-linolenic acid (20:3n-6, DGLA), AA, and adrenic acid (22:4n-6, AdA). Because the conversion of ALA and LA into long-chain PUFA depends on desaturase enzymes, we also examined the ratios of GLA to LA and of AA to DGLA as enzymatic activity indices for Δ6-desaturase (D6D) and Δ5-desaturase (D5D), respectively. Children's height-for-age *Z* scores were calculated according to the World Health Organization growth reference [32]. Maternal body mass index (BMI) was calculated as kg/m^2 from objectively measured height and weight in 35.7% of mothers and from self-reported data in the rest.

We first compared the distributions of total externalizing and internalizing problem scores across categories of baseline characteristics using means and standard deviation (SD). We then compared these behavior problem scores distributions across quartiles of FA concentrations. For ordinal exposures, we conducted tests for linear trend by fitting linear regression models with the behavior problem score as the continuous outcome and a variable representing ordinal categories of each predictor as a continuous covariate. For sex, we used the Wilcoxon rank-sum test. In adjusted analyses, we estimated mean differences with 95% confidence intervals (CIs) for total externalizing and internalizing problem scores between quartiles of FA. We also estimated unadjusted and adjusted mean differences with 95% CI in behavior problem scores per 1 SD of FA concentrations when the associations seemed linear. Adjustment covariates were baseline characteristics that were known independent predictors of behavior problems. These included child's sex, age, weekly hours spent watching television/playing video games, and maternal education and BMI. Prior literature suggests that girls exhibit higher levels of internalizing behavior problems, whereas boys have more externalizing behavior [33]. Prevalence of behavior problems in adolescence increases with age [34]. Time spent watching television or playing video games is associated with behavior problems in most studies [35]. Higher maternal education is related to less behavior problems in children [36]. Finally, maternal overweight and obesity may be associated with behavioral problems in the offspring [37]. Since PUFA may influence the development of adiposity [38], we did not consider child's BMI-for-age *Z* score as a covariate because this variable may be on the causal path between PUFA and behavior problems. In supplemental analyses, however, we adjusted for child's BMI-for-age *Z* score. In addition, we considered adjustment for all PUFA simultaneously. Since the number of participants with missing information in covariates was small (<10%), the primary multivariable analytic strategy consisted of complete case analyses. In additional sensitivity analyses, missing values in covariates were estimated using a Markov Chain Monte Carlo multiple imputation technique with ten imputation cycles before their inclusion in the multivariable models. We also examined the associations of PUFA with the subscales scores for total externalizing and internalizing problems following an analogous approach. Empirical estimates of the variance were specified in all models. All analyses were conducted with Statistical Analyses System version 9.4 (SAS Institute Inc.).

Results

Mean ± SD age at enrollment was 8.6 ± 1.6 years; 57.4% of children were girls. Mean ± SD ALA and LA concentrations were 0.5% and 30.6%, respectively (Supplemental Table 1). Serum FA were weakly to moderately correlated; the strongest correlations were between precursor FA and their products (Supplemental Table 2). Mean ± SD age at the time of follow-up assessment was 14.4 ± 1.6 years and mean ± SD total externalizing and internalizing problem scores were 52.0 ± 9.5 and 53.2 ± 9.7, respectively. Child's age at baseline and time spent watching television/playing video games were positively associated with total externalizing problem scores (Table 1). Child's BMI-for-age *Z* score and mother's BMI at baseline were positively related to total externalizing and internalizing problem scores, whereas mother's education level was inversely associated with these outcomes (Table 1).

Total externalizing problems

In bivariate analysis, EPA and DPA concentrations and the D5D activity index were inversely associated with total externalizing problem scores, whereas DHA concentration was positively related to this outcome (Table 2). In multivariable analysis, every SD of DPA or DHA concentration was related to a −0.9 ($P = 0.05$) and a 1.0 ($P = 0.03$) unit difference in total externalizing problem scores, respectively. Further adjustment for child's BMI-for-age *Z* score did not change these results (Supplemental Table 3). DHA remained positively associated with total externalizing problems after adjustment for all other PUFA; however, the inverse association between DPA and total externalizing problems became attenuated and not statistically significant

Table 1 Total behavior problem scores at 11–18 years of age according to sociodemographic characteristics in middle childhood among children from Bogota, Colombia

Characteristic	n[a]	Externalizing problems, mean ± SD	Internalizing problems, mean ± SD
Sex			
Boys	189	51.4 ± 9.5	52.4 ± 9.2
Girls	255	52.4 ± 9.4	53.8 ± 10.1
P[b]		0.25	0.13
Child's age at baseline, years			
5–6	78	48.5 ± 10.5	52.6 ± 11.5
7–8	171	51.1 ± 9.1	52.5 ± 9.2
9–10	170	53.7 ± 8.6	53.8 ± 9.2
11–12	25	56.5 ± 9.8	56.1 ± 11.0
P-trend[c]		<0.0001	0.12
Height-for-age Z score[d] at baseline			
<−2.0	35	51.4 ± 9.8	52.7 ± 9.4
−2.0 to <−1.0	135	51.0 ± 9.7	52.4 ± 10.1
−1.0 to <0.0	164	52.1 ± 9.2	53.8 ± 9.7
≥0.0	97	53.4 ± 9.4	53.8 ± 9.6
P-trend		0.08	0.24
BMI-for-age Z score[d] at baseline			
<−1.0	61	50.4 ± 8.9	51.2 ± 8.5
−1.0 to <0.0	145	51.0 ± 9.5	52.4 ± 10.3
0.0 to <1.0	152	53.0 ± 9.7	54.2 ± 9.5
≥1.0	73	53.2 ± 9.0	54.8 ± 10.0
P-trend		0.02	0.008
Time spent watching television/playing video games, h/week			
<10	140	51.1 ± 9.3	53.9 ± 8.7
10–<20	106	51.7 ± 9.9	52.4 ± 10.7
20–<30	115	51.5 ± 8.9	52.8 ± 10.0
≥30	76	54.7 ± 9.7	54.2 ± 9.8
P-trend		0.02	0.98
Mother's education, years			
Incomplete primary, 1–4	29	53.9 ± 8.5	56.4 ± 9.1
Complete primary, 5	82	53.3 ± 8.8	54.2 ± 10.0
Incomplete secondary, 6–10	105	51.9 ± 9.4	53.8 ± 9.8
Complete secondary, 11	189	51.4 ± 10.2	52.3 ± 9.7
University, >11	29	50.3 ± 8.1	50.8 ± 9.4
P-trend		0.03	0.005
Mother's height quartile (median), cm			
Q1, (150)	108	52.6 ± 9.8	54.8 ± 10.0
Q2, (155)	112	51.3 ± 10.0	52.0 ± 10.2
Q3, (160)	101	51.6 ± 8.9	53.8 ± 9.3
Q4, (165)	111	52.2 ± 9.3	52.4 ± 9.4
P-trend		0.79	0.20
Mother's BMI, kg/m^2			
<18.5	11	49.0 ± 8.9	47.2 ± 11.1
18.5–<25.0	259	51.5 ± 9.3	52.9 ± 9.7
25.0–<30.0	126	52.2 ± 9.8	53.4 ± 9.4
≥30.0	32	55.9 ± 9.5	57.6 ± 9.2
P-trend		0.02	0.007
Food insecurity			
Secure	108	51.3 ± 9.0	53.4 ± 9.6
Insecure—no hunger	222	51.9 ± 9.9	53.1 ± 10.0
Insecure—moderate hunger	61	53.2 ± 9.1	54.8 ± 9.1
Insecure—severe hunger	52	52.1 ± 9.3	51.8 ± 9.7
P-trend		0.35	0.66
Socioeconomic status			
1 (lowest)	33	49.7 ± 10.4	53.9 ± 10.8
2	101	51.9 ± 8.9	52.2 ± 9.9
3	250	52.6 ± 9.6	53.5 ± 9.7
4	60	50.6 ± 9.1	53.4 ± 9.1
P-trend		0.65	0.67

BMI body mass index

[a]Sums may be less than the total due to missing values in covariates

[b]Wilcoxon rank-sum test

[c]Test for linear trend when a variable representing ordinal categories of the characteristic was introduced into a linear regression model as a continuous covariate. Empirical estimates of the variance were used in all models

[d]According to the WHO growth reference for children and adolescents [32]

(Supplemental Table 4). Total externalizing problems were 3.2 units ($P = 0.02$) lower in children with a D5D activity index in the highest quartile compared with those in the lowest quartile (Table 2). Results did not change after multiple imputation of missing values (Supplemental Table 5).

Externalizing problems subscales

In multivariable analysis, DPA concentration and the D5D activity index were inversely related to aggressive behavior scores, whereas DHA concentration was positively associated with this outcome (Supplemental Table 6). DPA and AA concentrations and the D5D activity index were inversely associated with rule-breaking behavior scores (Supplemental Table 7). In addition, rule breaking behavior

Table 2 Total externalizing problem scores at 11–18 years of age according to serum FA percentage weight concentration in middle childhood among children from Bogota, Colombia

Total externalizing problem scores	Q1 $n = 111$	Q2 $n = 111$	Q3 $n = 111$	Q4 $n = 111$	P[a]	Mean difference per 1 SD[b]
n-3 polyunsaturated fatty acids (PUFA)						
18:3n-3 alpha-linolenic acid						
Mean ± SD	51.4 ± 8.8	51.7 ± 8.8	52.5 ± 10.4	52.3 ± 9.8	0.40	0.4 (−0.4, 1.3)
Adjusted differences[c]	Reference	0.4 (−2.0, 2.8)	0.3 (−2.2, 2.8)	0.9 (−1.5, 3.4)	0.48	0.4 (−0.4, 1.3)
20:5n-3 eicosapentaenoic acid						
Mean ± SD	53.0 ± 9.4	52.6 ± 9.7	51.4 ± 10.2	50.8 ± 8.4	0.05	−0.8 (−1.5, 0.0)
Adjusted differences	Reference	0.0 (−2.5, 2.4)	−0.1 (−2.6, 2.4)	−1.0 (−3.4, 1.3)	0.42	−0.3 (−1.1, 0.5)
22:5n-3 docosapentaenoic acid						
Mean ± SD	52.6 ± 10.0	52.3 ± 9.1	53.0 ± 9.3	49.8 ± 9.1	0.02	−1.0 (−1.8, −0.2)
Adjusted differences	Reference	−1.3 (−3.8, 1.2)	−0.2 (−2.7, 2.4)	−3.0 (−5.7, −0.3)	0.05	−0.9 (−1.7, 0.0)
22:6n-3 docosahexaenoic acid						
Mean ± SD	51.3 ± 9.7	51.7 ± 9.3	51.5 ± 9.9	53.3 ± 8.9	0.11	1.0 (0.1, 1.9)
Adjusted differences	Reference	0.9 (−1.5, 3.3)	−0.5 (−3.1, 2.1)	1.7 (−0.7, 4.1)	0.22	1.0 (0.1, 1.9)
n-6 PUFA						
18:2n-6 linoleic acid						
Mean ± SD	51.2 ± 9.7	51.6 ± 9.6	53.5 ± 9.6	51.6 ± 8.8	0.49	0.1 (−0.8, 0.9)
Adjusted differences	Reference	0.9 (−1.6, 3.4)	2.8 (0.3, 5.3)	0.6 (−1.8, 2.9)	0.41	0.2 (−0.7, 1.0)
18:3n-6 gamma-linolenic acid						
Mean ± SD	51.7 ± 9.3	52.2 ± 9.9	51.8 ± 9.5	52.1 ± 9.3	0.80	−0.1 (−1.0, 0.7)
Adjusted differences	Reference	−0.1 (−2.7, 2.4)	−0.5 (−3.0, 2.0)	−0.1 (−2.6, 2.5)	0.95	−0.1 (−1.0, 0.8)
20:2n-6 eicosadienoic acid						
Mean ± SD	52.2 ± 9.1	53.2 ± 10.2	51.4 ± 9.4	51.1 ± 9.0	0.23	−0.2 (−1.0, 0.6)
Adjusted differences	Reference	1.5 (−1.0, 4.1)	−0.9 (−3.3, 1.6)	−0.5 (−3.2, 2.1)	0.40	−0.1 (−1.0, 0.7)
20:3n-6 dihomo-gamma-linolenic acid						
Mean ± SD	50.5 ± 9.1	52.9 ± 10.0	52.0 ± 9.9	52.4 ± 8.8	0.20	0.3 (−0.6, 1.3)
Adjusted differences	Reference	2.4 (−0.1, 4.9)	1.1 (−1.4, 3.7)	2.6 (0.0, 5.3)	0.14	0.6 (−0.5, 1.7)
20:4n-6 arachidonic acid						
Mean ± SD	52.9 ± 9.7	52.3 ± 9.5	51.8 ± 9.1	50.8 ± 9.5	0.09	−0.9 (−1.8, 0.0)
Adjusted differences	Reference	0.2 (−2.4, 2.7)	−0.3 (−2.7, 2.1)	−1.8 (−4.3, 0.8)	0.15	−0.8 (−1.7, 0.1)
22:4n-6 adrenic acid						
Mean ± SD	52.8 ± 9.5	51.8 ± 9.0	51.8 ± 10.3	51.5 ± 9.1	0.28	−0.5 (−1.3, 0.4)
Adjusted differences	Reference	−1.6 (−3.9, 0.8)	−0.9 (−3.3, 1.6)	−1.6 (−4.0, 0.8)	0.25	−0.5 (−1.4, 0.3)
Enzyme activity indices						
Δ6-desaturase 18:3n-6/18:2n-6						
Mean ± SD	52.3 ± 9.3	51.3 ± 9.2	52.2 ± 10.0	52.1 ± 9.3	0.89	−0.2 (−1.0, 0.7)
Adjusted differences	Reference	−1.5 (−3.9, 0.9)	−0.4 (−2.9, 2.2)	−0.5 (−3.2, 2.2)	0.97	−0.2 (−1.2, 0.8)
Δ5-desaturase 20:4n-6/20:3n-6						
Mean ± SD	53.2 ± 9.7	51.1 ± 9.3	53.5 ± 9.4	50.0 ± 9.2	0.05	−0.8 (−1.8, 0.2)
Adjusted differences	Reference	−1.6 (−4.1, 0.8)	0.7 (−1.8, 3.2)	−3.2 (−5.8, −0.6)	0.05	−0.7 (−1.8, 0.3)

[a]Test for linear trend when a variable representing the median of each quartile was introduced into the linear regression model as a continuous predictor. Empirical estimates of the variance were specified in all models

[b]From a linear regression model with total externalizing problem score as the outcome and percentage of total serum FA per 1 SD (continuous) as the predictor

[c]Adjusted for child's sex (dichotomous), child's age at baseline (continuous), weekly hours spent watching television/playing video games (continuous), years of mother's education (continuous), and mother's body mass index (continuous). n-3 and n-6 PUFA were adjusted for the continuous percentage of total serum FA per 1 SD of linoleic acid and alpha-linolenic acid, respectively. Long-chain FA were additionally adjusted for their immediate FA precursor(s) (continuous). The Δ6-desaturase activity index was additionally adjusted for alpha-linolenic acid and linoleic acid (continuous). The Δ5-desaturase activity index was additionally adjusted for alpha-linolenic acid, gamma-linolenic acid, and eicosadienoic acid (continuous)

scores were higher in children with DGLA concentration in the highest quartile compared with those in the lowest quartile (Supplemental Table 7).

Total internalizing problems

In bivariate analysis, ALA concentration was positively associated with total internalizing problem scores, whereas AdA concentration was inversely associated with these scores (Table 3). After adjustment for potential confounders, every SD of ALA concentration was related to a 1.2 ($P = 0.01$) unit difference in total internalizing problem scores (Table 3). The adjusted difference in total internalizing problem scores between the highest and lowest quartile of AdA concentration was −3.2 units ($P = 0.01$) (Table 3). Further adjustment for child's BMI-for-age Z score did not change these results (Supplemental Table 8); however, after adjustment for other PUFA, the positive association between ALA and total internalizing problems was attenuated. AdA remained inversely related to this outcome (Supplemental Table 9). Results were not changed with multiple imputation of missing values (Supplemental Table 10).

Internalizing problems subscales

In multivariable analysis, ALA concentration was positively associated with anxious/depressed scores, whereas DPA and AdA concentrations and the D5D activity index were inversely related to this outcome (Supplemental Table 11). DPA concentration was inversely related to withdrawn/depressed scores in a nonlinear manner (Supplemental Table 12). AA concentration was also inversely associated with this outcome (Supplemental Table 12). There were no associations between PUFA concentrations and the somatic complaint scores (Supplemental Table 13).

Discussion

In this prospective study of schoolchildren from Bogotá, Colombia, higher serum concentrations of DHA in middle childhood were related to higher total externalizing problem scores in adolescence. The D5D enzyme activity index was inversely related to externalizing problems. ALA concentration was positively associated with internalizing problems, whereas AdA was inversely related to this outcome. These associations were independent of baseline child, parental, and household characteristics.

The positive association between serum DHA and total externalizing problem scores in adolescence was against our initial hypothesis. This association remained unchanged after additional adjustment for the child's BMI-for-age Z score and other PUFA. The majority of previous longitudinal studies have focused on the potential effects of DHA and other long-chain n-3 PUFA during the prenatal or infancy periods on early or middle childhood behavior problems. Only one [16] of the five [12–16] studies of DHA biomarkers during pregnancy found an association with childhood externalizing problems. In addition, maternal or early childhood intake of fatty fish, a major source of DHA, has not been related to behavior problems [39, 40]. DHA supplementation during the second half of pregnancy resulted in an increase of total problems at ages 4 and 7 years among Australian children [41, 42]. Only a handful of investigations have addressed the potential effect of DHA on behavior problems during adolescence. DHA supplementation of schoolchildren in Mauritius and the United Kingdom was related to decreased aggressive and disruptive behavior, respectively [18, 19]. The apparent discrepancy between the positive association we found and the protective effect of supplementation trials might be related to other PUFA present in the supplement. An adverse effect of DHA in middle childhood could be explained through a number of mechanisms. Fish oil supplementation has been associated with delayed neurodevelopment in rodents, potentially due to reduced myelination in the brainstem [43]. High DHA concentration has also been associated with behavioral delays in rodents when there is an imbalance of n-3 and n-6 PUFA [44]. Some methodological limitations of observational studies could also explain these findings. For example, the association of DHA and behavior problems in our study may be confounded by mercury or lead, since these neurotoxic heavy metals are readily found in fish consumed in Bogotá [45] and fish intake may be an important source of preformed DHA in this population.

The inverse associations of DPA with the internalizing problems subscales anxious/depressed and withdrawn/depressed have not been previously reported among children or adolescents. Nevertheless, DPA in blood has been inversely associated with depressive symptoms in pregnant [46] and postmenopausal women [47]. Evidence from animal experiments offers insights into potential explanatory mechanisms. Rodents supplemented with DPA perform better on the forced swim test [48], an animal model of depressive behavior. DPA could enhance hippocampal long-term potentiation [49], thereby countering stress- or glucocorticoid-induced damage that may be associated with neuropsychiatric disorders [50].

The D5D activity index in middle childhood was related to lower total externalizing problem scores in adolescence, possibly through decreased aggressive and rule-breaking behavior. Although no prior study has examined the association between the D5D activity index and behavior problems, some investigations have examined whether single-nucleotide polymorphisms that influence D5D activity are

Table 3 Total internalizing problem scores at 11–18 years of age according to serum fatty acid (FA) percentage weight concentration in middle childhood among children from Bogota, Colombia

Total internalizing problem scores	Q1 $n = 111$	Q2 $n = 111$	Q3 $n = 111$	Q4 $n = 111$	P[a]	Mean difference per 1 SD[b]
n-3 polyunsaturated fatty acids (PUFA)						
18:3n-3 alpha-linolenic acid						
Mean ± SD	52.3 ± 10.0	52.2 ± 9.4	53.5 ± 9.8	54.9 ± 9.6	0.03	1.0 (0.1, 2.0)
Adjusted differences[c]	Reference	0.0 (−2.7, 2.6)	1.1 (−1.6, 3.8)	2.8 (0.1, 5.5)	0.02	1.2 (0.3, 2.2)
20:5n-3 eicosapentaenoic acid						
Mean ± SD	52.5 ± 10.0	53.4 ± 9.6	53.7 ± 9.8	53.4 ± 9.5	0.47	0.2 (−0.7, 1.0)
Adjusted differences	Reference	1.1 (−1.4, 3.7)	1.7 (−0.9, 4.3)	1.5 (−1.1, 4.1)	0.22	0.4 (−0.4, 1.3)
22:5n-3 docosapentaenoic acid						
Mean ± SD	54.7 ± 11.2	53.3 ± 9.5	52.4 ± 9.1	52.5 ± 9.0	0.12	−0.4 (−1.3, 0.4)
Adjusted differences	Reference	−1.8 (−4.6, 1.0)	−2.6 (−5.3, 0.2)	−2.0 (−4.8, 0.9)	0.21	−0.2 (−1.1, 0.6)
22:6n-3 docosahexaenoic acid						
Mean ± SD	53.8 ± 10.5	53.2 ± 10.1	52.8 ± 9.1	53.1 ± 9.3	0.60	0.2 (−0.7, 1.2)
Adjusted differences	Reference	0.0 (−2.7, 2.7)	−0.9 (−3.5, 1.8)	−0.4 (−3.0, 2.2)	0.69	0.4 (−0.6, 1.3)
n-6 PUFA						
18:2n-6 linoleic acid						
Mean ± SD	52.3 ± 8.9	53.3 ± 9.9	54.1 ± 10.0	53.3 ± 10.1	0.37	0.3 (−0.6, 1.2)
Adjusted differences	Reference	0.7 (−1.8, 3.2)	1.2 (−1.3, 3.8)	0.7 (−1.8, 3.1)	0.54	0.2 (−0.6, 1.1)
18:3n-6 gamma-linolenic acid						
Mean ± SD	53.4 ± 10.0	53.4 ± 9.9	53.2 ± 9.5	52.9 ± 9.7	0.64	−0.3 (−1.2, 0.7)
Adjusted differences	Reference	−0.3 (−3.0, 2.3)	−0.4 (−3.1, 2.2)	−0.3 (−3.0, 2.4)	0.84	0.0 (−1.0, 1.0)
20:2n-6 eicosadienoic acid						
Mean ± SD	52.3 ± 9.8	54.5 ± 10.5	54.1 ± 9.7	52.1 ± 8.8	0.71	0.2 (−0.6, 1.1)
Adjusted differences	Reference	2.0 (−0.8, 4.8)	0.9 (−1.7, 3.5)	−0.5 (−3.2, 2.3)	0.58	0.2 (−0.8, 1.1)
20:3n-6 dihomo-gamma-linolenic acid						
Mean ± SD	52.4 ± 9.7	53.6 ± 10.1	53.9 ± 10.0	53.1 ± 9.2	0.57	0.0 (−0.9, 0.8)
Adjusted differences	Reference	1.1 (−1.6, 3.9)	1.2 (−1.6, 3.9)	0.9 (−2.0, 3.8)	0.60	−0.1 (−1.1, 0.9)
20:4n-6 arachidonic acid						
Mean ± SD	54.2 ± 9.7	53.4 ± 10.3	53.7 ± 9.4	51.8 ± 9.5	0.08	−0.8 (−1.6, 0.1)
Adjusted differences	Reference	−0.7 (−3.5, 2.0)	−0.3 (−2.8, 2.2)	−2.0 (−4.6, 0.5)	0.16	−0.6 (−1.5, 0.3)
22:4n-6 adrenic acid						
Mean ± SD	55.2 ± 10.3	52.4 ± 9.3	54.1 ± 9.8	51.3 ± 9.2	0.01	−1.1 (−2.1, −0.2)
Adjusted differences	Reference	−3.1 (−5.6, −0.6)	−0.3 (−2.9, 2.3)	−3.2 (−5.8, −0.7)	0.06	−0.8 (−1.7, 0.0)
Enzyme activity indices						
Δ6-desaturase 18:3n-6/18:2n-6						
Mean ± SD	54.1 ± 10.3	52.7 ± 9.6	53.5 ± 9.3	52.6 ± 9.8	0.40	−0.3 (−1.2, 0.5)
Adjusted differences	Reference	−1.1 (−3.8, 1.5)	−0.6 (−3.2, 2.1)	−0.9 (−3.8, 2.0)	0.67	−0.1 (−1.1, 0.9)
Δ5-desaturase 20:4n-6/20:3n-6						
Mean ± SD	53.7 ± 9.2	52.8 ± 10.2	55.1 ± 10.8	51.3 ± 8.4	0.14	−0.4 (−1.4, 0.6)
Adjusted differences	Reference	−0.4 (−3.0, 2.2)	1.8 (−0.8, 4.5)	−1.9 (−4.4, 0.5)	0.22	−0.2 (−1.1, 0.7)

[a]Test for linear trend when a variable representing the median of each quartile was introduced into the linear regression model as a continuous predictor. Empirical estimates of the variance were specified in all models

[b]From a linear regression model with total internalizing problem score as the outcome and percentage of total serum FA per 1 SD (continuous) as the predictor

[c]Adjusted for child's sex (dichotomous), child's age at baseline (continuous), weekly hours spent watching television/playing video games (continuous), years of mother's education (continuous) and mother's body mass index (continuous). n-3 and n-6 PUFA were adjusted for the continuous percentage of total serum FA per 1 SD of linoleic acid and alpha-linolenic acid, respectively. Long-chain FA were additionally adjusted for their immediate FA precursor(s) (continuous). The Δ6-desaturase activity index was additionally adjusted for alpha-linolenic acid and linoleic acid (continuous). The Δ5-desaturase activity index was additionally adjusted for alpha-linolenic acid, gamma-linolenic acid, and eicosadienoic acid (continuous)

related to child development. Single-nucleotide polymorphisms that encode for lower D5D activity were positively associated with infant psychomotor development [51] or child cognition [52], but it is unknown whether they are related to behavioral outcomes.

Serum ALA was positively related to total internalizing problem scores. These results are in line with those from a study of 10-year-old German children in whom prenatal ALA concentration was positively associated with peer relationship problems, an internalizing problem [13]. The nature of this association is unclear. Lifelong ALA supplementation has been associated with more depressive-like behaviors in rodents, potentially attributable to alterations in the endocannabinoid system [53]. Of note, the association became attenuated after adjustment for other PUFA.

AdA, a product of AA elongation, was inversely related to total internalizing problem scores, possibly through decreased anxious/depressed symptoms. AA was inversely related to problem scores in some subscales. Epidemiologic evidence of the potential effects of n-6 PUFA on behavior is limited and conflicting. Studies to date have only examined the association of prenatal or infant AA concentration with behavior problems. Three studies found no relation [12, 14, 15], one found an inverse association [13], and another found a positive relation [16]. Mechanisms underlying these associations are speculative; AA may enhance long-term potentiation of synapses in areas of the brain involved in reward [54], learning, and memory [55], while AdA could be important in myelination [56]. Of note, the D5D activity index and AA concentration were associated with rule-breaking behavior in the same direction. It is plausible that the associations with AA represent underlying genetic polymorphisms related to D5D activity.

EPA was not associated with any of the behavior problems we examined. These results are in contrast to a small trial in children [17] and to meta-analyses of randomized trials in adults with depressive behavior [20, 21]. There are several possible reasons for this discrepancy. First, the mechanisms of action in children may be different from those in adults. In addition, the majority of these trials were conducted among people with MDD, a rare condition in children. Therefore, results from these trials may not be generalizable to our population. Finally, in populations with low fish intake such as ours, EPA concentration may reflect differences in metabolism rather than differences in intake. Of note, we observed an inverse association between the D5D activity index and total externalizing problem scores and no associations between the D6D activity index and behavior problems. D5D participates in EPA synthesis, whereas D6D is on the path from DPA to DHA.

Our study has several strengths. First, the prospective design limits bias due to reverse causation. While previous studies primarily focused on DHA and AA, we examined the associations of other PUFA biomarkers that may be biologically relevant. Further, serum biomarkers of several FA are highly correlated with measures of long-term PUFA intake [57]. The YSR is a validated measure of externalizing and internalizing problems in adolescence [58]. Finally, we controlled for many potential confounders of the associations between PUFA concentration and behavior problems, including FA precursors. In this pediatric population, FA concentrations are related to age, BMI, socioeconomic status [38], and the type of cooking oil used in the household [59]. ALA is related to soybean oil intake, whereas long-chain n-3 and n-6 FA are related to soybean and sunflower intake. Whether interventions to improve the quality of dietary vegetable oils may be related to neurodevelopmental outcomes is a relevant future research direction.

There are limitations as well. We do not have a baseline measurement of behavior problems and thus cannot preclude reverse causation as an explanation for our findings if behavior problems developed before middle childhood. Our ability to identify a time at which PUFA may exert a potential effect on behavior is limited, since PUFA concentrations in middle childhood may be correlated with concentrations at other periods in development. Type I error may be enhanced by analyzing a large number of outcomes in relation to many individual biomarkers; thus we cannot rule out chance as an explanation for the associations observed. We were unable to measure heavy metals, which may share dietary sources with long-chain n-3 PUFA, especially EPA and DHA. Thus residual confounding may have attenuated or changed the direction of the associations. Also, we were unable to distinguish between active and passive screen time, which may exert differing effects on child behavior problems [60]. Finally, our results may not be generalizable to children from the highest socioeconomic status, since they do not typically attend public schools in Bogotá.

In conclusion, DHA concentration was positively associated with total externalizing problems. The D5D activity index was related to decreased externalizing problems. ALA concentration was positively associated with internalizing problems, whereas AdA was inversely related to this outcome. The associations of DHA and AdA with behavioral problems did not change after additional adjustment for other PUFA. Additional studies in other populations with different distributions of PUFA are warranted.

Acknowledgements We would like to thank the ASISA Foundation for providing funding.

Funding The study was supported by the ASISA Research Fund at the University of Michigan School of Public Health.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Publisher's note: Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

References

1. Hankin BL, Abramson LY, Moffitt TE, Silva PA, McGee R, Angel KE. Development of depression from preadolescent to young adulthood: emerging gender differences in a 10-year longitudinal study. J Abnorm Psychol. 1998;107:128–40.
2. Veldman K, Bultmann U, Almansa J, Reijneveld SA. Childhood adversities and educational attainment in young adulthood: the role of mental health problems in adolescence. J Adolesc Health. 2015;57:462–7.
3. Roza SJ, Hofstra MB, van der Ende J, Verhulst FC. Stable prediction of mood and anxiety disorders based on behavioral and emotional problems in childhood: a 14-year follow-up during childhood, adolescence, and young adulthood. Am J Psychiatry. 2003;160:2112–21.
4. Aebi M, Giger J, Plattner B, Metzke CW, Steinhausen HC. Problem coping skills, psychosocial adversities and mental health problems in children and adolescents as predictors of criminal outcomes in young adulthood. Eur Child Adolesc Psychiatry. 2014;23:283–93.
5. Martinez M. Tissue levels of polyunsaturated fatty acids during early human development. J Pediatr. 1992;120:S129–38.
6. McNamara RK, Carlson SE. Role of omega-3 fatty acids in brain development and function: potential implications for the pathogenesis and prevention of psychopathology. Prostaglandins Leukot Ess Fat Acids. 2006;75:329–49.
7. Davis PF, Ozias MK, Carlson SE, Reed GA, Winter MK, McCarson KE, et al. Dopamine receptor alterations in female rats with diet-induced decreased brain docosahexaenoic acid (DHA): interactions with reproductive status. Nutr Neurosci. 2010;13:161–9.
8. McNamara RK, Jandacek R, Rider T, Tso P, Cole-Strauss A, Lipton JW. Omega-3 fatty acid deficiency increases constitutive pro-inflammatory cytokine production in rats: relationship with central serotonin turnover. Prostaglandins Leukot Ess Fat Acids. 2010;83:185–91.
9. Deyama S, Ishikawa Y, Yoshikawa K, Shimoda K, Ide S, Satoh M, et al. Resolvin D1 and D2 reverse lipopolysaccharide-induced depression-like behaviors through the mTORC1 signaling pathway. Int J Neuropsychopharmacol. 2017;20:575–84.
10. Farooqui AA, Horrocks LA, Farooqui T. Modulation of inflammation in brain: a matter of fat. J Neurochem. 2007;101:577–99.
11. Kim YK, Na KS, Myint AM, Leonard BE. The role of pro-inflammatory cytokines in neuroinflammation, neurogenesis and the neuroendocrine system in major depression. Prog Neuropsychopharmacol Biol Psychiatry. 2016;64:277–84.
12. de Jong C, Kikkert HK, Seggers J, Boehm G, Decsi T, Hadders-Algra M. Neonatal fatty acid status and neurodevelopmental outcome at 9 years. Early Hum Dev. 2015;91:587–91.
13. Kohlboeck G, Glaser C, Tiesler C, Demmelmair H, Standl M, Romanos M, et al. Effect of fatty acid status in cord blood serum on children's behavioral difficulties at 10 y of age: results from the LISAplus Study. Am J Clin Nutr. 2011;94:1592–9.
14. Krabbendam L, Bakker E, Hornstra G, van Os J. Relationship between DHA status at birth and child problem behaviour at 7 years of age. Prostaglandins Leukot Ess Fat Acids. 2007;76:29–34.
15. Loomans EM, Van den Bergh BR, Schelling M, Vrijkotte TG, van Eijsden M. Maternal long-chain polyunsaturated fatty acid status during early pregnancy and children's risk of problem behavior at age 5-6 years. J Pediatr. 2014;164:762–8.
16. Steenweg-de Graaff JC, Tiemeier H, Basten MG, Rijlaarsdam J, Demmelmair H, Koletzko B, et al. Maternal LC-PUFA status during pregnancy and child problem behavior: the Generation R Study. Pedia Res. 2015;77:489–97.
17. Nemets H, Nemets B, Apter A, Bracha Z, Belmaker RH. Omega-3 treatment of childhood depression: a controlled, double-blind pilot study. Am J Psychiatry. 2006;163:1098–1100.
18. Tammam JD, Steinsaltz D, Bester DW, Semb-Andenaes T, Stein JF. A randomised double-blind placebo-controlled trial investigating the behavioural effects of vitamin, mineral and n-3 fatty acid supplementation in typically developing adolescent schoolchildren. Br J Nutr. 2016;115:361–73.
19. Raine A, Portnoy J, Liu J, Mahoomed T, Hibbeln JR. Reduction in behavior problems with omega-3 supplementation in children aged 8-16 years: a randomized, double-blind, placebo-controlled, stratified, parallel-group trial. J Child Psychol Psychiatry. 2015;56:509–20.
20. Mocking RJ, Harmsen I, Assies J, Koeter MW, Ruhe HG, Schene AH. Meta-analysis and meta-regression of omega-3 polyunsaturated fatty acid supplementation for major depressive disorder. Transl Psychiatry. 2016;6:e756.
21. Sublette ME, Ellis SP, Geant AL, Mann JJ. Meta-analysis of the effects of eicosapentaenoic acid (EPA) in clinical trials in depression. J Clin Psychiatry. 2011;72:1577–84.
22. Arsenault JE, Mora-Plazas M, Forero Y, Lopez-Arana S, Marin C, Baylin A, et al. Provision of a school snack is associated with vitamin B-12 status, linear growth, and morbidity in children from Bogota, Colombia. J Nutr. 2009;139:1744–50.
23. Harrison GG, Stormer A, Herman DR, Winham DM. Development of a Spanish-language version of the U.S. household food security survey module. J Nutr. 2003;133:1192–7.
24. Alvarez MC, Estrada A, Montoya EC, Melgar-Quiñonez H. Validation of a household food security scale in Antioquia, Colombia. Salud Publica Mex. 2006;48:474–81.
25. Robinson SL, Marín C, Oliveros H, Mora-Plazas M, Richards BJ, Lozoff B, et al. Iron Deficiency, anemia, and low vitamin B-12 serostatus in middle childhood are associated with behavior problems in adolescent boys: results from the Bogotá School Children Cohort. J Nutr. 2018;148:760–70.
26. Achenbach TM, Bird H, Canino G, Phares V, Gould M, Rubio-Stipec M. Epidemiological comparisons of Puerto Rican and U.S. mainland children: parent, teacher, and self-reports. J Am Acad Child Adolesc Psychiatry. 1990;29:84–93.
27. Achenbach TM, Rescorla LA. Reliability, internal consistency, cross-informant agreement, and stability. In: Achenbach TM, Rescorla LA, editors. Manual for the ASEBA school-age forms & profiles. (University of Vermont, Research Center for Children, Youth, & Families, Burlington, VT, 2001). p. 99–135.
28. Manual for the assessment data manager program (ADM) for the CBCL/4-18, YSR, TRF, YASR, YABCL, CBCL/2-3, CBCL/1½-5, & C-TRF, 2nd edn (University Medical Education Associates, Inc., Burlington, VT, 2000).
29. Corapci F, Calatroni A, Kaciroti N, Jimenez E, Lozoff B. Longitudinal evaluation of externalizing and internalizing behavior problems following iron deficiency in infancy. J Pedia Psychol. 2010;35:296–305.
30. Lozoff B, Castillo M, Clark KM, Smith JB, Sturza J. Iron supplementation in infancy contributes to more adaptive behavior at 10 years of age. J Nutr. 2014;144:838–45.

31. Morrison WR, Smith LM. Preparation of fatty acid methyl esters and dimethylacetals from lipids with boron fluoride-methanol. J Lipid Res. 1964;5:600–8.
32. de Onis M, Onyango AW, Borghi E, Siyam A, Nishida C, Siekmann J. Development of a WHO growth reference for school-aged children and adolescents. Bull World Health Organ. 2007;85:660–7.
33. Kramer MD, Krueger RF, Hicks BM. The role of internalizing and externalizing liability factors in accounting for gender differences in the prevalence of common psychopathological syndromes. Psychol Med. 2008;38:51–61.
34. Petersen IT, Bates JE, Dodge KA, Lansford JE, Pettit GS. Describing and predicting developmental profiles of externalizing problems from childhood to adulthood. Dev Psychopathol. 2015;27:791–818.
35. Suchert V, Hanewinkel R, Isensee B. Sedentary behavior and indicators of mental health in school-aged children and adolescents: a systematic review. Prev Med. 2015;76:48–57.
36. Carneiro P, Meghir C, Parey M. Maternal education, home environments, and the development of children and adolescents. J Eur Econ Assoc. 2013;11:123–60.
37. Bergmann S, Schlesier-Michel A, Wendt V, Grube M, Keitel-Korndorfer A, Gausche R, et al. Maternal weight predicts children's psychosocial development via parenting stress and emotional availability. Front Psychol. 2016;7:1156.
38. Perng W, Villamor E, Mora-Plazas M, Marin C, Baylin A. Alpha-linolenic acid (ALA) is inversely related to development of adiposity in school-age children. Eur J Clin Nutr. 2015;69:167–72.
39. Gale CR, Robinson SM, Godfrey KM, Law CM, Schlotz W, O'Callaghan FJ. Oily fish intake during pregnancy-association with lower hyperactivity but not with higher full-scale IQ in offspring. J Child Psychol Psychiatry. 2008;49:1061–8.
40. Waylen A, Ford T, Goodman R, Samara M, Wolke D. Can early intake of dietary omega-3 predict childhood externalizing behaviour? Acta Paediatr. 2009;98:1805–8.
41. Gould JF, Treyvaud K, Yelland LN, Anderson PJ, Smithers LG, McPhee AJ, et al. Seven-year follow-up of children born to women in a randomized trial of prenatal DHA supplementation. JAMA. 2017;317:1173–5.
42. Makrides M, Gould JF, Gawlik NR, Yelland LN, Smithers LG, Anderson PJ, et al. Four-year follow-up of children born to women in a randomized trial of prenatal DHA supplementation. JAMA. 2014;311:1802–4.
43. Saste MD, Carver JD, Stockard JE, Benford VJ, Chen LT, Phelps CP. Maternal diet fatty acid composition affects neurodevelopment in rat pups. J Nutr. 1998;128:740–3.
44. Wainwright PE, Jalali E, Mutsaers M, Bell R, Cvitkovic S. An imbalance of dietary essential fatty acids retards behavioral development in mice. Physiol Behav. 1999;66:833–9.
45. Lopez-Barrera EA, Barragan-Gonzalez RG. Metals and metalloid in eight fish species consumed by citizens of Bogota D.C., Colombia, and potential risk to humans. J Toxicol Environ Health A. 2016;79:232–43.
46. Pinto TJ, Vilela AA, Farias DR, Lepsch J, Cunha GM, Vaz JS, et al. Serum n-3 polyunsaturated fatty acids are inversely associated with longitudinal changes in depressive symptoms during pregnancy. Epidemiol Psychiatr Sci. 2017;26:157–68.
47. Jin Y, Kim TH, Park Y. Association between erythrocyte levels of n-3 polyunsaturated fatty acids and depression in postmenopausal women using or not using hormone therapy. Menopause. 2016;23:1012–8.
48. Laino CH, Garcia P, Podesta MF, Hocht C, Slobodianik N, Reines A. Fluoxetine potentiation of omega-3 fatty acid antidepressant effect: evaluating pharmacokinetic and brain fatty acid-related aspects in rodents. J Pharm Sci. 2014;103:3316–25.
49. Kelly L, Grehan B, Chiesa AD, O'Mara SM, Downer E, Sahyoun G, et al. The polyunsaturated fatty acids, EPA and DPA exert a protective effect in the hippocampus of the aged rat. Neurobiol Aging. 2011;32:2318 e2311–5.
50. Park HJ, Lee S, Jung JW, Kim BC, Ryu JH, Kim DH. Glucocorticoid- and long-term stress-induced aberrant synaptic plasticity are mediated by activation of the glucocorticoid receptor. Arch Pharm Res. 2015;38:1204–12.
51. Yeates AJ, Love TM, Engstrom K, Mulhern MS, McSorley EM, Grzesik K, et al. Genetic variation in FADS genes is associated with maternal long-chain PUFA status but not with cognitive development of infants in a high fish-eating observational study. Prostaglandins Leukot Ess Fat Acids. 2015;102–103:13–20.
52. Steer CD, Lattka E, Koletzko B, Golding J, Hibbeln JR. Maternal fatty acids in pregnancy, FADS polymorphisms, and child intelligence quotient at 8 y of age. Am J Clin Nutr. 2013;98:1575–82.
53. Zamberletti E, Piscitelli F, De Castro V, Murru E, Gabaglio M, Colucci P, et al. Lifelong imbalanced LA/ALA intake impairs emotional and cognitive behavior via changes in brain endocannabinoid system. J Lipid Res. 2017;58:301–16.
54. Dumuis A, Pin JP, Oomagari K, Sebben M, Bockaert J. Arachidonic acid released from striatal neurons by joint stimulation of ionotropic and matabotropic quisqualate receptors. Nature. 1990;347:182–4.
55. McGahon B, Clements MP, Lynch MA. The ability of aged rats to sustain long-term potentiation is restored when the age-related decrease in membrane arachidonic acid concentration is reversed. Neuroscience. 1997;81:9–16.
56. Wijendran V, Lawrence P, Diau G, Boehm G, Nathanielsz PW, Brenna JT. Significant utilization of dietary arachidonic acid is for brain adrenic acid in baboon neonates. J Lipid Res. 2002;43:762–7.
57. Baylin A, Kim MK, Donovan-Palmer A, Siles X, Dougherty L, Tocco P, et al. Fasting whole blood as a biomarker of essential fatty acid intake in epidemiologic studies: comparison with adipose tissue and plasma. Am J Epidemiol. 2005;162:373–81.
58. Ivanova MY, Achenbach TM, Rescorla LA, Dumenci L, Almqvist F, Bilenberg N, et al. The generalizability of the Youth Self-Report syndrome structure in 23 societies. J Consult Clin Psychol. 2007;75:729–38.
59. Villamor E, Marin C, Mora-Plazas M, Casale M, Vargas LN, Baylin A. Cooking with soyabean oil increases whole-blood alpha-linolenic acid in school-aged children: results from a randomized trial. Public Health Nutr. 2015;18:3420–8.
60. Sweetser P, Johnson D, Ozdowska A, Wyeth P. Active versus passive screen time for young children. Aust J Early Child. 2012;37:94–8.