ORIGINAL RESEARCH

CURRENT DEVELOPMENTS IN NUTRITION

Nutrition in Health and Disease

Higher n–6:n–3 Fatty Acid Intake Is Associated with Decreased Cardiometabolic Risk Factors in a Racially Diverse Sample of Children

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Abstract

Background: Accumulating evidence implicates diet quality in childhood as playing a significant role in adult cardiometabolic health. Polyunsaturated fatty acids (PUFAs) of the n–6 (ω-6) and n–3 (ω-3) series contribute unique protective effects against cardiometabolic disease. As such, the ratio between n–6 and n–3 PUFAs is a dietary metric of interest in the early life span, although an optimum intake ratio has yet to be determined.

Objective: This cross-sectional study assesses relations between the ratio of total n–6:n–3 PUFA intake and cardiometabolic risk factors in a racially diverse sample of children ($n = 191$) from the Admixture Mapping of Ethnic and Racial Insulin Complex Outcomes (AMERICO) study.

Methods: Outcome measures included waist circumference, lipid concentrations, fasting glucose, and two 24-h dietary recalls from boys and girls aged 7–12 y who self-reported as European American ($n = 81$), African American ($n = 55$), or Hispanic American ($n = 55$). Linear regression analyses were used to assess associations between predictors of interest and outcomes after adjusting for covariates.

Results: PUFA intake reflected in the n–6:n–3 ratio was inversely associated with concentrations of total and LDL cholesterol [$\beta \pm$ SE: -0.359 ± 0.107 ($P = 0.001$) and -0.189 ± 0.069 ($P = 0.007$), respectively]. Exploratory analyses showed that the intake of total n–6 PUFAs was not significantly predictive of any cardiometabolic risk factor assessed, whereas total n–3 PUFA intake was positively associated with concentrations of HDL cholesterol ($\beta \pm$ SE: 0.114 ± 0.042; $P = 0.007$).

Conclusions: Results suggest that the effect of n–6 and n–3 PUFA intake reflected in the ratio may be largely driven by n–3 PUFAs in reducing 2 lipid cardiometabolic risk factors among this multiethnic cohort of children. Until an ideal intake ratio is determined, nutritional counseling should focus on meeting recommended levels of both n–3 and n–6 PUFAs in order to establish beneficial childhood dietary patterns that may positively influence adult cardiometabolic health. *Curr Dev Nutr* 2018;2:nzy014.

Introduction

It is well established that dietary FAs have differing effects on cardiometabolic risk factors such as serum lipids, insulin sensitivity, and body composition (1–3). Such findings have shaped the Dietary Guidelines for Americans, which emphasize decreased intake of *trans* FAs and SFAs because the intakes of both are negatively associated with cardiometabolic health (4). In contrast to the deleterious effects of *trans* FAs and SFAs, PUFAs of the n–6 and n–3 series exhibit unique protective effects against cardiometabolic disease. For example, n–6 PUFAs exert direct action on circulating lipid concentrations, whereas n–3 PUFAs influence overall cardiac function (5). In addition, n–6 and n–3 PUFAs are metabolized to yield eicosanoids, which elicit physiologic effects influencing metabolic health, including inflammation and insulin sensitivity (6, 7).

Keywords: polyunsaturated fatty acids, children, cardiometabolic disease risk, dietary patterns

Manuscript received July 18, 2017. Initial review completed November 29, 2017. Revision accepted February 28, 2018. Published online April 3, 2018.

The authors reported no funding received for this study.

Author disclosures: KMC-W, MIC, HHB, TH, and JRF, no conflicts of interest.

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Abbreviations used: AFADM, African admixture; AI, Adequate Intake; ALA, α-linolenic acid; AMERICO, Admixture Mapping of Ethnic and Racial Insulin Complex Outcomes; LA, linoleic acid; MVPA, moderate to vigorous physical activity; NDSR, Nutrient Data System for Research; SES, socioeconomic status; TC, total cholesterol; UAB, University of Alabama at Birmingham; WC, waist circumference.

Although the manifestation of cardiometabolic disease typically occurs in adulthood, dietary patterns contributing to disease are often formed in childhood. A systematic review of 20 studies with a dietary follow-up of 6–27 y from baseline showed accumulating evidence that childhood nutrition and dietary patterns play a significant role in adult cardiovascular health (8). Furthermore, it is widely recognized that dietary patterns established in youth often persist in adulthood (9–11). In light of these findings, early assessment of dietary behaviors in childhood may be paramount in guiding the establishment of a healthy diet in order to reduce the risk of developing cardiometabolic disease.

The National Academy of Medicine Health and Medicine Division has established age-recommended Adequate Intake (AI) levels for the essential n–6 fatty acid, linoleic acid [LA, 18:2 (n–6)], and for α-linolenic acid [ALA, 18:3 (n–3)], the essential n–3 PUFA (12). Beyond individual intake, it has been proposed that the intake ratio of n–6 to n–3 PUFAs may also influence cardiometabolic health. For example, evidence from prospective cohort studies and secondary-prevention trials suggest that a close balance of the ratio may have protective cardiometabolic effects (13–15); however, an optimum intake ratio has yet to be determined. Thus, the aim of this study was to assess associations between cardiometabolic risk factors with dietary intakes of total n–6 and total n–3 FAs and the total n–6:n–3 among a racially diverse sample of children, comparing overall intake with AI recommended levels according to sex and age. Cardiometabolic risk factors evaluated in the study include waist circumference (WC), HDL cholesterol, LDL cholesterol, total cholesterol (TC), TGs, and fasting glucose.

Methods

Subjects

The current study uses cross-sectional data from the previously described Admixture Mapping of Ethnic and Racial Insulin Complex Outcomes (AMERICO) study (NIH R01 DK067426), which collected detailed genetic, dietary, behavioral, and metabolic data from a multiethnic cohort of healthy children in the United States (16). In short, participants ($n = 191$) included in this ancillary study were between the ages of 7 and 12 y, including self-reported European American ($n = 81$), African American ($n = 55$), and Hispanic American ($n = 55$) boys and girls recruited in Birmingham, Alabama. Although the AMERICO study included 311 children, this ancillary study included only 191 children due to missing data in the variables of interest. All of the participants were classified by a pediatrician as peri- or prepubertal, which was defined by a score of ≤ 3 with the use of criteria developed by Marshall and Tanner (17, 18). No participants with a medical diagnosis, previous medical condition, or medication that might influence cardiometabolic health or body composition were included in the AMERICO study. Both the AMERICO study and the ancillary study described herein were approved by the Institutional Review Board at the University of Alabama at Birmingham (UAB) and the Institutional Review Board at the University of Alabama.

Dietary intake

Two 24-h dietary recalls conducted on 2 different weekdays were completed, with each child using the multiple-pass method of the Nutrient Data System for Research (NDSR; Nutrition Coordinating Center, University of Minnesota). Recalls were performed with ≥ 1 parent present in order to determine that the reported intake reflected typical intake. Intake data were coded and entered into NDSR version 6 by a registered dietitian. The 2 recalls were averaged to obtain PUFA intake data. Total n–3 PUFA intake reflects the sum of ALA, EPA [20:5 (n–3)], and DHA [22:6 (n–3)] and total n–6 PUFA intake is the sum of LA and arachidonic acid. The n–6 to n–3 PUFA ratio was calculated by using total n–6 PUFA and total n–3 PUFA intakes.

Anthropometric measurements and biochemical assessments

WC was measured by the study dietitian at the narrowest part of the torso or the area between the ribs and iliac crest with the use of a flexible tape measure (GullicI II; Country Technologi, Inc.) and recorded to the nearest 0.1 cm (19). Fasting blood samples were collected at 0700 after an overnight stay at the UAB General Clinical Research Center. Concentrations of all serum-derived analytes were measured at the UAB Metabolism Core Laboratory. Glucose was measured in 10-μL serum samples using an Ektachem DT System (Johnson and Johnson Clinical Diagnostics). The intra-assay CV for this analysis was 0.61%, and the mean interassay CV was 1.45%. Lipids were measured by using a Stanbio SIRRUS analyzer. LDL cholesterol was calculated with the use of the Friedewald method (20).

Physical activity assessment

It is well documented that physical activity is inversely associated with weight, blood lipids, and fasting glucose (21). To control for this influence on variables of interest, participants wore an MTI ActiGraph accelerometer for 7 d (GT1M; ActiGraph Health Services), with acceptable removal of the device at times of sleep or water exposure only. Epoch length was set at 1 min, and activity was expressed as counts per minute. Moderate to vigorous physical activity (MVPA) was defined as >1952 counts/min (22).

Socioeconomic status

Socioeconomic status (SES) was assessed because it may influence dietary intake and has been associated with body composition in children (23). The Hollingshead 4-factor index of social status was administered because it combines educational and occupational status into a score ranging from 8 to 66 (24).

Genetic admixture

Estimates of African (AFADM), Amerindian, and European admixture were used to account for the biodiversity within and among racial/ethnic categories and for the potential confounding of ancestral genetic contribution to cardiometabolic outcomes in children. Genetic admixture estimates were calculated through maximum likelihood estimation (25) with the use of genotyping from ~142 ancestry informative markers across the human genome, as described elsewhere (26). This measure estimates the proportion of genetic ancestry for an individual by using a range of proportions from 0 to 1 and identifies the most probable value of admixture on the basis of the observed genotypes.

Statistical analysis

All of the analyses were conducted with the use of SAS 9.4 (SAS Institute). Demographic and outcome variables were summarized as

means ± SDs for continuous variables or as percentages (*n*) for categorical variables. Outcome variables that were non-normally distributed were transformed to meet the model assumption. Comparisons of absolute values among racial/ethnic classification were performed by ANOVA for continuous variables or chi-square tests for frequencies. WC (centimeters) was reciprocal transformed. HDL cholesterol, LDL cholesterol, and TG concentrations (millimoles per liter) were $\log_2$-transformed; thus, the regression coefficients can be interpreted as doubling the rate of the lipid concentrations. TC was kept in its original unit as millimoles per liter. Energy intake was divided by 100 to improve computing precision by the software. Exploratory analyses were used to identify the most parsimonious models to explain the outcomes of interest while taking into consideration the role of statistics and physiologic factors. A general linear regression model was used to assess the association between the independent variables and the outcomes, adjusting for covariates including sex, Tanner stage, SES, AFADM, European admixture, and MVPA. The model for WC was also adjusted for height. Power calculations informed that the addition of 1 more predictor of interest to a model with 9 covariates (the most complex model tested), given a sample size of 191 and an $\alpha = 0.01$, provided 81–85% power to detect a difference in R^2 of 0.06, within a range of full-model R^2 of 0.12–0.20, and 82% power to detect a difference in R^2 of 0.04 for a full-model $R^2 = 0.42$. Residual analyses were conducted for each outcome to check independence and normality assumptions using

TABLE 1 Demographic characteristics and metabolic risk factors[1]

	Total (*n* = 191)	EA (*n* = 81)	AA (*n* = 55)	HA (*n* = 55)	*P*
Demographic characteristics					0.53
Age, y	9.61 ± 1.53	9.67 ± 1.60	9.69 ± 1.45	9.41 ± 1.51	
Sex, % (*n*)					0.82
Male	53 (101)	51 (41)	56 (31)	53 (29)	
Female	47 (90)	49 (40)	44 (24)	47 (26)	
BMI,[2] kg/m^2	18.49 ± 2.95	17.83 ± 2.58	18.11 ± 3.07	19.83 ± 2.97	<0.0001
Underweight, % (*n*)	0.52 (1)	1 (1)	0 (0)	0 (0)	
Normal weight, % (*n*)	69 (132)	77 (62)	76 (42)	51 (28)	
Overweight, % (*n*)	19 (37)	15 (12)	15 (8)	29 (16)	
Obese, % (*n*)	12 (22)	7 (6)	9 (5)	20 (11)	
Height, cm	140 ± 10	140 ± 10	142 ± 11	137 ± 10	0.06
Weight, kg	36.43 ± 8.94	35.08 ± 8.13	36.95 ± 9.44	37.92 ± 9.44	0.18
PUFA intake[3]					
Total n–6 PUFAs, g	12.15 ± 5.99	11.09 ± 4.21	14.80 ± 8.21	11.06 ± 4.75	0.006
Total n–3 PUFAs, g	1.25 ± 0.69	1.12 ± 0.54	1.46 ± 0.90	1.23 ± 0.60	0.07
n–6:n–3	10.46 ± 3.53	10.84 ± 3.72	10.84 ± 3.66	9.52 ± 2.96	0.02
Total PUFAs, g	13.49 ± 6.55	12.28 ± 4.59	16.37 ± 8.94	12.39 ± 5.30	0.006
Linoleic acid, g	12.04 ± 5.96	11.01 ± 4.19	14.67 ± 8.17	10.93 ± 4.72	0.006
α-Linolenic acid, g	1.14 ± 0.61	1.02 ± 0.44	1.32 ± 0.79	1.15 ± 0.58	0.09
Arachidonic acid, g	0.11 ± 0.09	0.09 ± 0.06	0.13 ± 0.13	0.13 ± 0.07	0.0007
EPA+DHA, g	0.09 ± 0.26	0.09 ± 0.21	0.12 ± 0.39	0.07 ± 0.10	0.06
Metabolic risk factors					
WC, cm	64.39 ± 8.83	62.87 ± 7.32	61.99 ± 8.16	69.01 ± 9.86	<0.0001
HDL-C, mmol/L	1.28 ± 0.31	1.25 ± 0.29	1.39 ± 0.34	1.21 ± 0.30	0.006
LDL-C, mmol/L	2.30 ± 0.66	2.34 ± 0.58	2.21 ± 0.67	2.32 ± 0.75	0.38
TC, mmol/L	3.94 ± 0.68	3.94 ± 0.60	3.90 ± 0.68	3.98 ± 0.78	0.91
TGs, mmol/L	1.77 ± 0.97	1.72 ± 0.77	1.44 ± 0.80	2.17 ± 1.24	<0.0001
Fasting glucose, mmol/L	2.53 ± 0.17	2.51 ± 0.15	2.47 ± 0.16	2.61 ± 0.16	0.0002
Total energy intake, kcal	1903 ± 444	1844 ± 348	1975 ± 512	1918 ± 491	0.41
MVPA	55.43 ± 29.19	59.65 ± 29.41	54.88 ± 30.26	49.78 ± 27.22	0.18
Tanner score, % (*n*)					
Boys					0.10
1	70 (71)	76 (31)	58 (18)	76 (22)	
2	21 (21)	22 (9)	23 (7)	17 (5)	
3	8 (8)	2 (1)	19 (6)	3.5 (1)	
4	1 (1)	0 (0)	0 (0)	3.5 (1)	
Girls					0.19
1	58 (52)	68 (27)	37.5 (9)	61.5 (16)	
2	27 (24)	20 (8)	37.5 (9)	27 (7)	
3	16 (14)	12 (5)	25 (6)	11.5 (3)	
4	0 (0)	0 (0)	0 (0)	0 (0)	

[1]Demographic and outcome variables are means ± SDs. Continuous variables were compared by the Kruskal-Wallis nonparametric test, and categorical variables were compared by using Fisher's exact test. AA, African American; EA, European American; HA, Hispanic American; HDL-C, HDL cholesterol; LDL-C, LDL cholesterol; MVPA, moderate to vigorous physical activity; TC, total cholesterol; WC, waist circumference.
[2]Results are based on a BMI percentile calculator for children.
[3]Total n–3 PUFA intake is the sum of α-linolenic acid, EPA, and DHA and total n–6 PUFA intake is the sum of linoleic acid and arachidonic acid. The n–6-to-n–3 PUFA ratio was calculated by using the total n–6 PUFA and total n–3 PUFA intakes.

TABLE 2 Results of general linear model analyses for cardiometabolic risk factors[1]

	Associations with n–6:n–3											
	WC		HDL-C		LDL-C		TC		TGs		Glucose	
Variable	$\beta \pm$ SE	*P*	$\beta \pm$ SE	*P*	$\beta \pm$ SE	*P*	$\beta \pm$ SE	*P*	$\beta \pm$ SE	*P*	$\beta \pm$ SE	*P*
AFADM	0.371 ± 0.207	0.075	1.034 ± 0.469	0.029	−1.223 ± 0.586	0.038	−1.183 ± 0.918	0.200	−0.775 ± 0.831	0.352	−0.270 ± 0.224	0.229
EUADM	0.046 ± 0.109	0.674	0.238 ± 0.247	0.335	0.359 ± 0.308	0.246	0.675 ± 0.483	0.164	−0.513 ± 0.437	0.242	−0.194 ± 0.118	0.101
Race												
AA	−0.094 ± 0.156	0.548	−0.451 ± 0.352	0.202	1.024 ± 0.441	0.021	1.135 ± 0.690	0.102	−0.131 ± 0.624	0.834	0.029 ± 0.168	0.862
EA	0.090 ± 0.064	0.165	0.016 ± 0.145	0.912	−0.166 ± 0.182	0.363	−0.358 ± 0.285	0.210	0.063 ± 0.258	0.806	0.057 ± 0.069	0.416
MVPA	0.001 ± 0.000	0.002	0.001 ± 0.001	0.492	−0.002 ± 0.001	0.079	−0.003 ± 0.002	0.066	−0.002 ± 0.002	0.125	−0.001 ± 0.000	0.127
n–6:n–3	0.015 ± 0.024	0.525	−0.093 ± 0.055	0.093	−0.189 ± 0.069	0.007	−0.359 ± 0.107	0.001	−0.020 ± 0.097	0.835	−0.006 ± 0.026	0.805

[1] $\beta \pm$ SE. All models were adjusted for sex, Tanner stage, and socioeconomic status. "AMIND" is the reference group for admixture comparisons and Hispanics are the reference group for comparison between race/ethnicity groups. MVPA was defined as >1952 counts/min. WC (cm) was reciprocal transformed. HDL-C, LDL-C, and TG concentrations (mmol/L) as well as n–6:n–3 were $\log_2$-transformed; thus, the regression coefficients can be interpreted as doubling the rate of the lipid concentrations. AA, African American; AFADM, African admixture; AMIND, Amerindian admixture; EA, European American; EUADM, European admixture; HDL-C, HDL cholesterol; LDL-C, LDL cholesterol; MVPA, moderate to vigorous physical activity; TC, total cholesterol; WC, waist circumference.

the Kolmogorov-Smirnov test. The parameter estimates, SEs, and corresponding *P* values were reported. To account for the potential effect of multiple comparisons, a conservative significance level ($P \leq 0.01$) was chosen to control for type I error.

Results

Descriptive statistics and metabolic risk factors are reported in **Table 1**. African-American children reported significantly greater total n–6 PUFA intake, total PUFA intake, and intake of LA ($P = 0.006$ for all intake variables). The intake ratio of n–6:n–3 was significantly lower among Hispanic-American children ($P = 0.02$).

The relation between n–6:n–3 PUFA intake and cardiometabolic outcomes is outlined in **Table 2**. Results suggest that n–6:n–3 was inversely associated with LDL-cholesterol and TC concentrations ($P = 0.007$ and 0.0010, respectively) after adjusting for covariates. AFADM was significantly associated with higher LDL-cholesterol concentrations in this cohort ($P < 0.021$). In exploratory analyses, the intake of total n–6 PUFAs was not significantly predictive of any cardiometabolic risk factor assessed in this study, whereas total n–3 PUFA intake was positively associated with HDL-cholesterol concentrations ($P = 0.007$) after adjusting for covariates. Furthermore, children of higher AFADM had significantly higher HDL-cholesterol concentrations ($P < 0.029$), and children of later pubertal status showed significantly lower HDL-cholesterol concentrations ($P = 0.008$). In addition, European-American children had significantly lower LDL-cholesterol concentrations compared with African-American children ($P = 0.01$) after adjusting for sex, pubertal stage, SES, and admixture. Total PUFA intake did not significantly predict TG or glucose concentrations after adjusting for covariates, although children with later pubertal status had significantly lower TG concentrations ($P = 0.006$).

Comparative assessments of ALA and LA intakes with AI levels for life stage and sex are reported in **Table 3**. Results indicate that 46.9% of boys and 59.2% of girls met the recommendation for LA intake, whereas 46% of boys and 54.9% of girls met the AI for ALA intake.

Discussion

Results of this study suggest that a higher intake of total n–3 PUFAs is associated with higher HDL cholesterol, whereas a higher n–6:n–3 PUFA intake is associated with significant reductions in TC and LDL cholesterol in this cohort of healthy children. Results between higher n–3 PUFA intake and higher n–6:n–3 are likely attributable to the unique mechanistic functions of n–6 and n–3 PUFAs in relation to cardiometabolic health. For example, PUFAs of the n–6 series exert direct action on reducing TC as well as LDL cholesterol, an established risk factor for cardiovascular disease (5). In contrast, protective mechanisms of n–3 PUFAs may be derived from their ability to lower TGs and attenuate inflammation, thereby improving glucose metabolism (27–29). Although no PUFA intake measure was predictive of WC, previously reported results suggest that ratio of n–6 to n–3 was negatively associated with intra-abdominal adipose tissue ($P = 0.014$), a more sensitive measurement of abdominal adiposity than WC (30). In comparison to AI levels, LA and ALA intakes in this cohort were similar to other studies reporting PUFA intake in this demographic group to be below recommended levels (31, 32). In addition, there were some significant associations between the covariates considered in the study and the cardiometabolic outcomes, including one between AFADM and LDL cholesterol and another between MVPA and WC. These associations are not unexpected findings, given previously reported data on this cohort that support relations between ancestral genetics and lipid profiles as well as metabolic outcomes with physical activity (33, 34).

TABLE 3 Percentage of children meeting AI recommendations specific to life stage and sex[1]

	Meeting AI, %
Linoleic acid	
Boys	
Age 4–8 y: 10 g/d	53.8
Age 9–13 y: 12 g/d	40.0
Girls	
Age 4–8 y: 10 g/d	57.6
Age 9–13 y: 10 g/d	60.9
Alpha-linolenic acid	
Boys	
Age 4–8 y: 0.9 g/d	61.5
Age 9–13 y: 1.2 g/d	30.6
Girls	
Age 4–8 y: 0.9 g/d	61.5
Age 9–13 y: 1.0 g/d	48.4

[1] Results are based on AI recommendations by the National Academy of Medicine Health and Medicine Division. AI, Adequate Intake.

Although the results reflect an inverse relation between the n–6 to n–3 ratio and TC as well as LDL cholesterol, it must be acknowledged that a diet high in n–6 PUFAs has been called into question for its potential to blunt the cardioprotective effects of n–3 PUFAs (6, 35, 36). Thus, it has been proposed that a more balanced n–6 to n–3 PUFA ratio may maximize therapeutic benefit to be derived from each PUFA class. Presently, the relation between n–6:n–3 and cardiometabolic disease remains poorly characterized, with results often confounded by the multifactorial nature of chronic disease. As such, it has been proposed that the ratio may vary by specific risk factors and disease states (35); furthermore, the ratio may vary between adults and children, given the intense developmental processes of childhood.

Despite the lack of consistent evidence to support an optimal ratio in adults and children, results from the National Heart, Lung, and Blood Institute Family Heart Study, conducted in >4500 participants with a mean age of 52 y, suggest that a higher intake of either LA or ALA was inversely and independently associated with a prevalence OR of cardiovascular disease (28); however, when the intake of both was in the highest tertile, there was a 56% lower prevalence OR than for participants in the lower tertile of either FA intake. Taken collectively, meeting the recommended intakes of LA and ALA may assist in reducing cardiometabolic disease risk, especially until an optimum ratio is established.

Our study provides insightful results, but it is not exempt from some limitations. Despite the use of the validated multi-pass method of the NDSR to reduce dietary-reporting inaccuracies and conducting recalls with a trained dietitian in the presence of a parent in order to ensure reporting accuracy, our results are limited by the use of self-reported dietary intake. In addition, there was no assessment of circulating blood PUFA concentrations, although previous research suggests that PUFA concentrations are fairly well correlated with dietary intake assessments (37, 38).

Strengths of the study include the well-represented cohort of multiethnic children along with robust analyses that accounted for several covariates known to influence dietary intake and metabolic markers (sex, SES, genetic admixture, pubertal stage, etc.). However, it is important to remember that the cross-sectional design of this study precludes the ability to ascribe causality both due to potential confounding and a lack of knowledge about the temporal relation between variables of interest.

Results of this study suggest an interactive effect between n–6 and n–3 PUFAs reflected in the n–6 to n–3 ratio on decreasing 2 lipid cardiometabolic risk factors among this ethnically diverse sample of children; however, the intake of n–6 PUFAs was not associated with any cardiometabolic risk factor and the intake of n–3 PUFAs was significantly associated with higher HDL cholesterol. In light of these results, it is plausible to consider that the predictive results observed in the ratio may be mediated by n–3 intake. Overall, intake amounts of both LA and ALA fell far short of AI recommendations for life stage and sex. Because an optimal intake ratio of n–6 to n–3 has yet to be determined, nutritional counseling for optimizing cardiometabolic health in children should focus on meeting recommended levels for PUFA intake. Such nutritional counseling is critical to establishing beneficial childhood dietary patterns that may positively influence adult cardiometabolic health.

Acknowledgments

The authors' responsibilities were as follows—KMC-W, MIC, and JRF: designed the research; KMC-W and HHB: conducted the research and wrote the manuscript; TH and JRF: analyzed the data; and all authors: contributed substantially to the editing of the manuscript and read and approved the final manuscript.

References

1. Food and Agriculture Organization of the United Nations. Fats and fatty acids in human nutrition: report of an expert consultation. www.fao.org/3/a-i1953e.pdf 2010 . Accessed March 4, 2017.
2. Astrup A, Dyerberg J, Elwood P, Hermansen K, Hu FB, Jakobsen MU, Kok FJ, Krauss RM, Lecerf JM, LeGrand P, et al. The role of reducing intakes of saturated fat in the prevention of cardiovascular disease: where does the evidence stand in 2010? Am J Clin Nutr 2011;93:684–8.
3. Lopez-Garcia E, Schulze M, Manson JE, Meigs JB, Albert CM, Rifai N, Willett WC, Hu FB. Consumption of (n-3) fatty acids is related to plasma biomarkers of inflammation and endothelial activation in women. J Nutr 2004;134:1806–11.
4. US Department of Health and Human Services; USDA. Dietary guidelines for Americans, 2015–2020. 8th ed. Dec 2015. [cited 2016 Jan 29]. https://health.gov/dietaryguidelines/2015/.
5. Wijendran V, Hayes KC. Dietary n-6 and fatty acid balance and cardiovascular health. Annu Rev Nutr 2004;24:597–615.
6. Patterson E, Wall R, Fitzgerald GF, Ross RP, Stanton C. Health implications of high dietary omega-6 polyunsaturated fatty acids. J Nutr Metab 2012; 2012:16
7. Lorente-Cebrian S, Costa AGV, Navas-Carretero S, Zabala M, Martinez JA, Moreno-Aliaga MJ. Role of omega-3 fatty acids in obesity, metabolic syndrome, and cardiovascular diseases: a review of the evidence. J Physiol Biochem 2013;69:633–51.
8. Kaikkonen JE, Mikkila V, Raitakari OT. Role of childhood food patterns on adult cardiovascular disease risk. Curr Atheroscler Rep 2014;16:443.
9. Wang Y, Bentley ME, Zhai F, Popkin BM. Tracking of dietary intake patterns of Chinese from childhood to adolescence over a six-year follow up period. J Nutr 2002;132:430–8.
10. Lien N, Lytle LA, Klepp KI. Stability in consumption of fruit, vegetables, and sugary foods in a cohort from age 14 to age 21. Prev Med 2001;33:217–26.
11. Mikkila V, Rasanen L, Raitakari OT, Pietinen P, Viikari J. Consistent dietary patterns identified from childhood to adulthood: the Cardiovascular Risk in Young Finns Study. Br J Nutr 2005;93:923–31.
12. National Academy of Sciences. Dietary Reference Intakes for energy, carbohydrate, fiber, fat, fatty acids, cholesterol, protein, and amino acids and dietary reference intakes for water, potassium, sodium, chloride, and sulfate. 2005. [cited 2016 Jan 29]. Available from: www.nap.edu.
13. Bucher H, Hengstler P, Schindler C, Meier G. N-3 polyunsaturated fatty acids in coronary heart disease: a meta-analysis of randomized controlled trials. Am J Med 2002;112:298–304.
14. Hu FB, Bronner L, Willett WC, Stampfer MJ, Rexrod KM, Albert CM, Hunter D, Manson JE. Fish and omega-3 fatty acid intake and risk of human thrombosis and hemostasis. Am J Clin Nutr 2002;65:1687–98.
15. Griffin M, Sanders T, Davies IG, Morgan LM, Millward DJ, Lewis F, Slaughter S, Cooper JA, Miller GJ, Griffin BA. Effects of altering the ratio of dietary n-6 to n-3 fatty acids on insulin sensitivity, lipoprotein size, postprandial lipemia in men and postmenopausal women aged 45–70 y: the OPTILIP study. Am J Clin Nutr 2006;84:1290–8.
16. Cardel M, Higgins PB, Willig AL, Keita AD, Casazza K, Gower BA, Fernandez JR. African genetic admixture is associated with body composition and fat distribution in a cross-sectional study of children. Int J Obes (Lond) 2011;35:60–5.

17. Marshall WA, Tanner JM. Variations in pattern of pubertal changes in girls. Arch Dis Child 1969;44:291–303.
18. Marshall WA, Tanner JM. Variations in pattern of pubertal changes in boys. Arch Dis Child 1970;45:13–23.
19. Lohman TG, Going SB. Body composition assessment for development of an international growth standard for preadolescent and adolescent children. Food Nutr Bull 2006;27:S314–25.
20. Friedewald WT, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma without use of an ultracentrifuge. Clin Chem 1972;18:499–502.
21. US Department of Health and Human Services Physical Activity Guidelines Advisory Committee. Physical activity guidelines advisory committee report. 2008. [cited 2016 Jan 29]. Available from: https://health.gov/paguidelines/Report/pdf/CommitteeReport.pdf.
22. Willig AL, Hunter GR, Casazza K, Heimburger DC, Beasley TM, Fernandez JR. Body fat and racial genetic admixture are associated with aerobic fitness levels in a multiethnic pediatric population. Obesity (Silver Spring) 2011;19:222–7.
23. Cardel M, Willig AL, Dulin-Keita A, Casazza K, Beasley TM, Fernandez JR. Parental feeding practices and socioeconomic status are associated with child adiposity in a multi-ethnic sample of children. Appetite 2012;58:347–53.
24. Hollingshead A. Four factor index of social status. New Haven (CT): Yale University Press; 1975.
25. Hanis CL, Chakraborty R, Ferrell RE, Schull WJ. Individual admixture estimates: disease associations and individual risk of diabetes and gallbladder disease among Mexican-Americans in Starr County, Texas. Am J Phys Anthropol 1986;70:433–41.
26. Klimentidis YC, Divers J, Casazza K, Beasley TM, Allison DB, Fernandez JR. Ancestry-informative markers on chromosome 2, 8 and 15 are associated with insulin-related traits in a racially diverse sample of children. Hum Genomics 2011;5:79–89.
27. Roche HM, Gibney MJ. Effect of long-chain n-3 polyunsaturated fatty acids on fasting and postprandial triacylglyerol metabolism. Am J Clin Nutr 2000;71(Suppl):232S–7S.
28. Djousse L, Pankow JS, Folsom AR, Hopkins PN, Province MA, Hong Y, Ellison RC. Relation between dietary linolenic acid and coronary artery disease in the National Heart, Lung, and Blood Institute Family Heart Study. Am J Clin Nutr 2001;74:612–9.
29. Marzi C, Huth C, Baumert J, Thorand B, Rathmann W, Meisinger C, Wichmann HE, Roden M, Peters A, Grallert H, et al. Acute-phase serum amyloid A protein and its implication in the development of type 2 diabetes in the KORA S4/F4 study. Diabetes Care 2013;36:1321–6.
30. Cardel M, Lemas DJ, Jackson KH, Friedman JE, Fernandez JR. Higher intake of PUFAs is associated with lower total and visceral adiposity and higher lean mass in a racially diverse sample of children. J Nutr 2015;145:2146–52.
31. Harika RK, Cosgrove MC, Osendarp SJM, Verhoef P, Zock PL. Fatty acid intakes are not in line with the dietary intake recommendations for future cardiovascular health: a systematic review of dietary intake data from thirty countries. Br J Nutr 2011;106:307–16.
32. Madden S, Garrioch C, Holub B. Direct diet quantification indicates low intakes of (n-3) fatty acids in children 4 to 8 years old. J Nutr 2009;139:528–32.
33. Cassazza K, Willig AL, Gower BA, Nagy TR, Hunter GR, Wallace S, Amaya M, Franklin F, Beasley M, Fernandez JR. The role of European genetic admixture in the etiology of the insulin resistance syndrome in children: are the effects mediated by fat accumulation? J Pediatr 2010;157(1):50–56.
34. Casazza K, Gower BA, Willig AL, Hunter GR, Fernandez JR. Physical fitness, activity, and insulin dynamics in early pubertal children. Pediatr Exerc Sci 2009;21:63–76.
35. Simopoulos AP. The importance of the ratio of omega-6/omega-3 essential fatty acids. Biomed Pharmacother 2002;56:365–79.
36. Simopoulos AP. The importance of the omega-6/omega-3 fatty acid ratio in cardiovascular disease and other chronic diseases. Exp Biol Med (Maywood) 2008;233:674–88.
37. Hodson L, Skeaff C, Fielding B. Fatty acid composition of adipose tissue and blood in humans and its use as a biomarker of dietary intake. Prog Lipid Res 2008;47:348–80.
38. Browning L, Walker C, Mander AP, West AL, Madden J, Gambell JM, Young S, Wang L, Jebb SA, Calder PC. Incorporation of eicosapentaenoic and docosahexaenoic acids into lipid pools when given as supplements providing doses equivalent to typical intakes of oily fish. Am J Clin Nutr 2012;96:748–58.