

Concurrent Physical Activity Modifies the Association between n3 Long-Chain Fatty Acids and Cardiometabolic Risk in Midlife Adults^{1,2}

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Abstract

Greater consumption of n3 (ω 3) polyunsaturated fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) can reduce risk for cardiovascular disease events, yet their effects on metabolic risk factors and diabetes remain unclear. This cross-sectional study used a community volunteer sample to test whether the associations between n3 fatty acids and cardiometabolic risk vary as a function of physical activity. Participants were 344 generally healthy adults, 30–54 y of age, not taking fish oil supplements or confounding medications. Serum phospholipid EPA and DHA were used together (EPA+DHA) as a biomarker of n3 fatty acid exposure. Cardiometabolic risk was calculated as a continuous measure based on standardized distributions of blood pressure, waist circumference, HDL cholesterol, triglycerides, glucose, and a simple count of risk factors. Insulin resistance was estimated from the homeostatic model assessment. Physical activity was found to predict cardiometabolic risk ($P \leq 0.02$) and insulin resistance ($P \leq 0.02$) and to moderate the association between EPA+DHA and both cardiometabolic risk (P -interaction ≤ 0.02) and insulin resistance (P -interaction ≤ 0.02). Specifically, higher EPA+DHA was associated with lower cardiometabolic risk and insulin resistance in persons engaged in regular physical activity but not in relatively inactive individuals. These findings were noted in several components of cardiometabolic risk, in men and women separately, and in models adjusted for overall diet quality. In midlife adults, habitual physical activity may be necessary to unmask the salutary effects of n3 fatty acids on cardiometabolic risk and insulin resistance. *J. Nutr.* 143: 1414–1420, 2013.

Introduction

Fish consumption appears to reduce major cardiovascular disease events (1). This benefit is thought to derive principally from the n3, long-chain (LC-¹⁰) PUFAs, EPA and DHA, nutrients found almost exclusively in seafood. EPA and DHA have pleiotropic effects on cellular functioning, and the biologic mechanism(s) of their health effects are multifaceted. In part, the cardiovascular disease protection provided by EPA and DHA may occur through salutary effects on insulin resistance, correlated cardiometabolic risk factors that comprise metabolic syndrome (MetSyn), and type II diabetes. Experiments in animals (2) and epidemiologic observations (3,4) have found greater EPA and DHA consumption to be related to greater insulin sensitivity

and a lower incidence of diabetes. However, other reports have not associated low fish intake with diabetes or its precursor, MetSyn (5,6), and have even found high EPA and DHA intake to be associated with an elevated incidence of diabetes or comparatively high serum glucose concentrations (7–9).

Regular physical activity is another positive health behavior that prevents cardiovascular disease. The benefits of exercise may accrue through blood pressure (BP) reduction, as suggested by short-term experiments and prospective epidemiologic investigations (10,11). Aerobic physical activity also mitigates insulin resistance and related lipid abnormalities, which also could contribute to cardiovascular disease prevention (12,13).

Given the overlap in the putative cardiometabolic effects of EPA and DHA consumption and regular physical activity, we examined the associations of both health behaviors with cardiometabolic risk and insulin resistance in a single investigation, specifically to test the hypothesis that effects of n3 LC-PUFA exposure vary as a function of habitual physical activity. The molecular biomediators of the effects of EPA and DHA are, to a certain extent, antagonized by biomediators derived from

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¹⁰ Abbreviations used: AA, arachidonic acid; BP, blood pressure; HDL-C, HDL cholesterol; LC, long-chain; MetSyn, metabolic syndrome.

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arachidonic acid (AA), an n6 fatty acid. Therefore, the ratio of n6/n3 fatty acids was also examined in relation to cardiometabolic risk.

Participants and Methods

Participants. This study used data from a convenience subsample of the Adult Health and Behavioral project, a cross-sectional study completed between 2001 and 2005. The parent project comprised behavioral and biologic information collected from generally healthy volunteers between the ages of 30 and 54 y living in the Pittsburgh metropolitan area. Volunteers were recruited through mass mailing of study information, and a total of 1379 participants completed the parent study. Exclusion criteria included 1) clinically evident atherosclerotic vascular disease and diabetes (determined by fasting glucose and diabetic medication use); 2) known liver or kidney disease, cancer, or major neuropsychiatric disorder; and 3) antihypertensive, lipid-lowering, or psychotropic medication use. Those who completed the parent study after December 2001 ($n = 1046$) were asked to participate in further studies that included, among other measures, blood samples for fatty acid analysis and dietary interviews, if they met the following additional criteria: 1) no fish oil supplement use, 2) resting BP <180/110 mm Hg, 3) body mass index (weight/height²) <40 kg/m², and 4) mean weekly alcohol consumption ≤ 21 drinks per week (ethanol <273 g/wk). Some participants (287) did not meet these additional criteria, and some (396) declined to participate. The sample size of 363 was further reduced to 344 because various technical difficulties caused a lack of serum fatty acid results for 19 participants. The study protocol was approved by the University of Pittsburgh's Institutional Review Board (no. 0805006 and 000535), and informed consent was obtained from all participants in accordance with university Institutional Review Board guidelines.

Cardiometabolic measures. Before attending a morning appointment, participants were asked to fast for 8 h and avoid exercise for 12 h and alcohol for 24 h. During the appointment, a nurse completed a medical history and medication use interview; obtained height, weight, and waist-circumference measurements; and drew a 40-mL blood sample. For BP and pulse measurement, participants sat quietly for at least 5 min and arm circumference was measured to determine the appropriate cuff size. Two BP readings were auscultated with a mercury manometer at 2-min intervals by certified staff; between intervals, pulse rate was palpated for 30 s. Systolic and diastolic BP readings were averaged. Mean BP was calculated from diastolic BP + pulse pressure/3.

Serum glucose, HDL cholesterol (HDL-C), and triglyceride concentrations were measured by the Heinz Nutrition Laboratory (School of Public Health, University of Pittsburgh), which has met criteria for the Centers for Disease Control and Prevention–National Heart, Lung, and Blood Institute Standardization Program since 1982. Serum insulin concentration was measured in duplicate with a RIA (Code-a-count; Diagnostic Products). An estimate of insulin resistance was calculated as follows: HOMA-IR = serum insulin ($\mu\text{IU/mL}$) \times fasting blood glucose (mmol/L)/22.5 (14).

Whereas MetSyn is defined as present or absent on the basis of threshold values for each of 5 risk factors (15), use of cardiometabolic risk as a continuous measure better predicts future cardiovascular disease events (16). For that reason, a composite, continuous index of cardiometabolic risk was calculated using unit weighting of the 5 MetSyn criteria (15): BP, waist circumference, fasting HDL-C, triglycerides, and glucose.

Serum phospholipid fatty acid composition. A fasting sample of whole blood was centrifuged to separate RBCs from protein-rich serum. Phospholipids were extracted and separated from the serum samples by Sep-Pak (Waters) silica cartridges. Fatty acid methyl esters were prepared from serum phospholipids using methanolic KOH reagent. And diheptadecanoyl lecithin (Matraya) was used as an internal standard. The capillary GC method used to determine levels of serum phospholipid fatty acids was essentially the same as described in another study (17). Peaks on the chromatograms were identified by comparing the retention

times with those of standard mixtures (Supelco) and were calculated by a ChemStation (Revision A.09.03; Agilent Technologies), using an internal standard mode. Intra- and interassay coefficients of variation were 2.0–9.2% and 1.9–9.6%, respectively, for all major serum fatty acids. Individual fatty acids were expressed as percent of total fatty acids (mol%). Serum phospholipid EPA and DHA were summed to generate a measure of circulating long-chain n3 fatty acid content. An index of the ratio of n6/n3 LC-PUFAs was calculated as follows: AA/(EPA+DHA).

Physical activity. Physical activity was assessed using the Paffenbarger physical activity questionnaire (18). This instrument queries an individual regarding activities in the past year expressed as the daily distance walked, pace of walking, and flights of stairs climbed as well as type, duration, and frequency of exercise, sports, and other recreational activities. Activities are categorized by rate of energy expenditure, from which weekly total expenditures are estimated. The mean value in healthy adult populations is generally 2000–2500 kcal/wk, but the value is generally regarded as an indicator of physical activity rather than a measure of true energy expenditure. The questionnaire has good test-retest reliability ($r = 0.72$), has reasonable validity in comparison with other physical activity surveys, and predicts risk for myocardial infarction and all-cause mortality (18,19).

Diet. Two, unannounced 24-h diet recall interviews were conducted with each participant by telephone. The interviews used the Nutrition Data System for Research, a Windows-based dietary analysis program designed for the collection and analyses of 24-h dietary recalls (Nutrition Coordination Center at the University of Minnesota, <http://www.ncc.umn.edu/>) (20). Consumption of fiber, folate, sodium, and saturated fat were expressed per 2000 kcal and used as indicators of diet quality.

Statistical analysis. All statistical analyses were performed using SPSS (Version 19.0, SPSS). Race of the participants was categorized into 2 groups: Caucasians ($n = 304$) and other ($n = 40$). We transformed continuous variables with a skewness >1.5 using logarithm base 10 to normalize distributions.

For derivation of a continuous cardiometabolic risk score, the 5 MetSyn criteria (BP, waist circumference, fasting HDL-C, triglycerides, and glucose) were each standardized. HDL-C was reverse scored. The 5 measures were summed, and the resulting distribution was restandardized.

Multivariate linear regression models were created using age, race, and sex as covariates; fatty acid indices and physical activity as independent variables; and cardiometabolic risk score and insulin resistance as dependent variables. Additional regression models were constructed that included the interaction between serum fatty acids and physical activity. The interaction term was calculated as the arithmetic product of mean-centered fatty acids and physical activity. Analyses were conducted among all participants and among men and women separately.

Statistically significant interactions between fatty acids and exercise were explored by constructing linear regression models of logged physical activity (1 SD above and 1 SD below the mean). The regression coefficient for EPA+DHA in each model was examined for statistical significance and used to graph the findings. The Mann–Whitney U test was used to compare groups of participants with respect to the number of metabolic criteria risk factors. An alpha level of $P < 0.05$ was interpreted as statistically significant.

Results

Characteristics of study participants were recorded (Table 1). These participants met criteria that excluded persons with diabetes, any vascular disease, and severe hypertension or obesity, and they volunteered to participate. Compared with excluded individuals, study participants were similar in age, gender, HDL-C, triglycerides, and physical activity, but the majority were Caucasian and had more education and higher BMI, BP,

TABLE 1 Characteristics of study participants¹

Characteristic	Male (n = 161)	Female (n = 183)
Race, % Caucasian	90	87
Age, y	44.3 ± 6.8	44.7 ± 6.6
Education, % with bachelor's degree	60	61
Body mass index, kg/m	27.2 ± 4.2	25.5 ± 4.5*
Waist circumference, cm	97 ± 12	83 ± 12*
Systolic BP, mm Hg	120 ± 12	111 ± 12*
Diastolic BP, mm Hg	81 ± 9	75 ± 8
HDL-C, mg/dL	47.3 ± 10.8	60.2 ± 14.8
Triglycerides, mg/dL	113 (75–170)	83 (64–113)
Glucose, mg/dL	96.4 ± 10.0	91.9 ± 9.6*
Insulin resistance (HOMA-IR)	2.78 (2.05–4.06)	2.38 (1.78–3.27)*
Physical activity, kcal/wk	2100 (1170–3180)	2070 (1110–3330)
Serum phospholipid EPA+DHA, mol%	1.91 (1.51–2.49)	2.05 (1.61–2.77)
Serum phospholipid AA/(EPA+DHA), mol%/mol%	4.70 ± 1.76	4.40 ± 1.64
Dietary sodium ² , mg/d	4500 ± 1700	2900 ± 1100*
Saturated fat, g/d	35 ± 15	23 ± 10*
Fiber, g/d	21 ± 10	16 ± 7*
Folate, mg/d	610 ± 280	420 ± 190*

¹ Mean ± SD or median (IQR). These data were log-transformed for statistical analysis. *Different from males, $P < 0.05$. AA, arachidonic acid; BP, blood pressure; HDL-C, HDL cholesterol.

² Data available on 138 males and 153 females.

glucose, and insulin resistance. An example of the study sample's median physical activity level is a participant who reported walking 12 blocks and climbing 10 sets of stairs daily during the course of routine activities, walking for exercise 45 min 5 times a week, and coaching baseball practice for 1 h 10 times a year.

The cardiometabolic risk score consisted of the 5 risk factors equally weighted: BP, waist circumference, HDL-C, fasting triglycerides, and fasting glucose. The distribution of the cardiometabolic risk score was mean ± SD = 0 ± 1. Cardiometabolic risk was higher in men (0.56 ± 0.88) than in women (−0.49 ± 0.82) ($P < 0.001$) but was unrelated to age in this sample. In simple bivariate analyses, cardiometabolic risk correlated inversely with physical activity ($r = -0.18$, $P < 0.05$) and EPA+DHA ($r = -0.13$, $P < 0.05$), and directly with the ratio of n6/n3 PUFAs [i.e., AA/(EPA+DHA)] ($r = 0.16$, $P < 0.05$).

We obtained results from multiple linear regression analyses (Table 2). Adjusted for age, sex, and race, physical activity was related to lower cardiometabolic risk, whereas EPA+DHA was not (model 1). However, a significant interaction was observed between EPA+DHA and physical activity (model 2), that is, higher EPA+DHA was associated with lower cardiometabolic

risk, specifically in persons with relatively high levels of physical activity. We illustrate this finding by separately modeling the association between EPA+DHA and cardiometabolic risk in subjects with relatively low and relatively high physical activity (Fig. 1A). In high exercisers (1 SD above the mean), higher EPA+DHA exposure was associated with lower cardiometabolic risk ($P < 0.001$). At physical activity 1 SD below the mean, greater EPA+DHA tended to be associated with greater cardiometabolic risk ($P = 0.07$).

Similarly, EPA+DHA interacted with physical activity in regression models of insulin resistance at high and low levels of physical activity (Table 2). Analogous to the results for cardiometabolic risk, physical activity moderated the association between EPA+DHA and insulin resistance (Fig. 1B). In high exercisers, higher EPA+DHA exposure was associated with lower insulin resistance ($P = 0.04$), whereas among low exercisers, EPA+DHA was unrelated to insulin resistance.

Regression analyses based on the ratio of n6/n3 polyunsaturated fatty acids [AA/(EPA+DHA)] yielded comparable results, albeit with reversed sign as expected (Table 2). AA/(EPA+DHA) was related to both cardiometabolic risk score and insulin

TABLE 2 Linear regression analyses of n3 LC-PUFAs and physical activity as predictors of cardiometabolic risk score and insulin resistance in midlife adults¹

Model ²	Variable	Cardiometabolic risk score			Insulin resistance		
		R ²	β (95% CI)	P	R ²	β (95% CI)	P
1	EPA+DHA	0.32	−0.06 (−0.15, 0.03)	0.19	0.06	−0.01 (−0.12, 0.10)	0.86
	Physical activity		−0.16 (−0.26, −0.07)	0.001		−0.13 (−0.24, −0.02)	0.02
2	Interaction	0.34	−0.16 (−0.25, −0.07)	<0.001	0.08	−0.14 (−0.25, −0.04)	0.007
1	AA/(EPA+DHA)	0.32	.06 (−0.04, 0.15)	0.23	0.06	0.00 (−0.11, 0.11)	0.98
	Physical activity		−0.17 (−0.26, −0.07)	<0.001		−0.13 (−0.24, −0.03)	0.02
2	Interaction	0.34	0.17 (0.09, 0.26)	<0.001	0.09	0.17 (0.06, 0.27)	0.002

¹ AA, arachidonic acid; LC-PUFA, long-chain, polyunsaturated fatty acid.

² Covariates are age, race, and sex in both models.

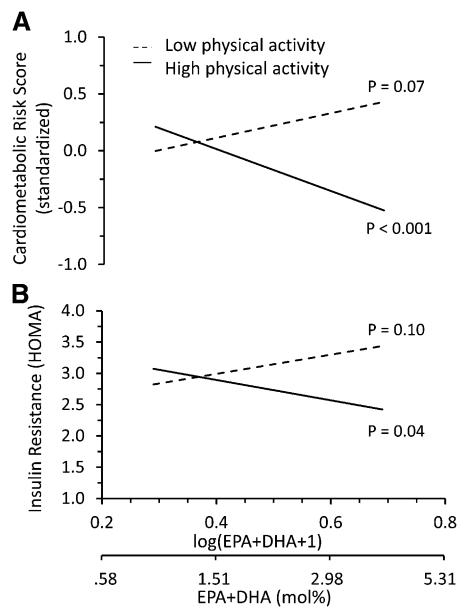


FIGURE 1 Cardiometabolic risk score (A) and insulin resistance (B) in midlife adults as a function of eicosapentaenoic acid + docosahexaenoic acid (EPA+DHA) content in fasting serum phospholipids. (A) The association between cardiometabolic risk and EPA+DHA varied significantly as a function of physical activity. Higher EPA+DHA was associated with lower cardiometabolic risk when modeled on relatively high physical activity, defined as 1 SD above the mean (4100 kcal/wk). In contrast, higher EPA+DHA in persons with lower physical activity (modeled at 1 SD below the mean, ~900 kcal/wk) tended to be associated with higher cardiometabolic risk. (B) The association between insulin resistance and EPA+DHA similarly varied as a function of physical activity. Covariates included age, gender, and race.

resistance in interaction with physical activity. Specifically, among high exercisers a higher AA/(EPA+DHA) ratio was associated with a higher cardiometabolic risk score and insulin resistance (data not shown).

To illustrate these findings in terms of clinical MetSyn criteria, the number of risk factors was examined in participants grouped by low or high physical activity and low or high EPA+DHA (each categorized by median split). Participants high in both physical activity and EPA+DHA had significantly fewer MetSyn risk factors than the other 3 groups ($P = 0.004$) (Fig. 2).

In sex-stratified multivariate linear analyses of cardiometabolic risk, the interaction between physical activity and EPA+DHA was significant in men ($P = 0.03$) and women ($P = 0.004$) as was the sex-stratified interaction between physical activity and AA/(EPA+DHA) in men ($P = 0.005$) and women ($P = 0.004$). In sex-stratified regression models of insulin resistance, marginally significant interactions were found between physical activity and EPA+DHA in men ($P = 0.07$) and women ($P = 0.06$). In addition, significant interactions were found between physical activity and AA/(EPA+DHA) in men ($P = 0.02$) and women ($P = 0.05$).

Analyses were repeated in the 291 participants with available dietary data to control for the potentially confounding effects of overall diet quality. Four indicators of diet quality (fiber, folate, sodium, and saturated fat) were added as covariates, each expressed per 2000 kcal. All the interaction terms remained statistically significant (Table 2).

We studied individual cardiometabolic risk factors (Table 3). In model 1 (before inclusion of the interaction term), higher EPA+DHA levels were not associated with risk factors, whereas

greater physical activity was related to higher HDL and lower waist circumference, triglycerides, and, at a trend level, glucose ($P = 0.07$). In model 2, EPA+DHA interacted with exercise in predicting waist circumference, HDL, triglycerides, and, at a trend level, BP ($P = 0.06$). Analyses based on the AA/(EPA+DHA) ratio yielded similar associations.

Discussion

The current analyses were spurred by contradictory results of research on n3 LC-PUFA in relation to insulin resistance and cardiometabolic risk. The clustered indicators comprising cardiometabolic risk represent a complex phenotype with multiple determinants. Therefore, the effects of dietary fatty acid consumption on cardiometabolic risk may vary as a function of other contributing factors. In the current sample of 344 midlife, generally healthy community volunteers, the associations between serum phospholipid n3 fatty acid content and both cardiometabolic risk and insulin resistance were moderated by habitual physical activity. EPA+DHA exposure was associated favorably with cardiometabolic risk and insulin resistance in persons reporting relatively high levels of physical activity but not otherwise. This set of findings was also analyzed on the basis of the number of MetSyn criteria met and was duplicated using the ratio of n6/n3 LC-PUFAs. These interactions were found in men and women separately, persisted with additional adjustments for overall diet quality, and extended to individual components of cardiometabolic risk.

Consumption of fish or supplementation with fish oil has been associated with lower rates of cardiovascular disease in epidemiologic studies and in some, but not all, randomized clinical trials (1,21). The n3 fatty acids plentiful in fish, EPA, and DHA reliably lower serum triglyceride concentration but manifest a range of other, less definitive, mechanisms of action. Laboratory studies have indicated that these micronutrients serve as precursors of anti-inflammatory compounds, quench reactive oxygen species, serve as ligands for peroxisome proliferator-activated receptor (PPAR) transcription factors, and prevent the development of insulin resistance (2,5,22). To the extent that diabetes develops from mismanagement of chronic fuel surfeit with resultant toxic lipid infiltration of myocytes, hepatocytes, and pancreatic islet cells (23), the ability of EPA and DHA to

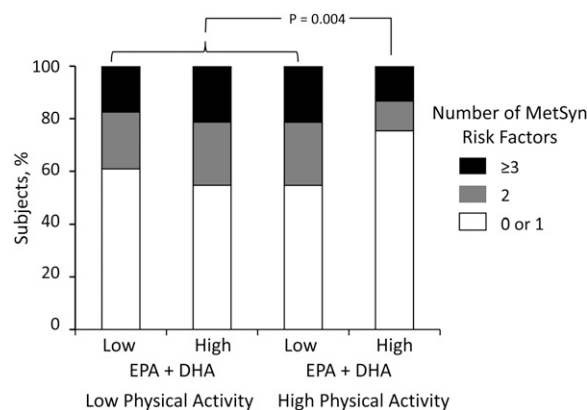


FIGURE 2 Number of metabolic syndrome (MetSyn) risk factors in midlife adults grouped according to low and high physical activity and low and high eicosapentaenoic acid + docosahexaenoic acid (EPA+DHA) in serum phospholipids. Groups based on median split of physical activity (<2093 vs. ≥2093 kcal/wk) and EPA+DHA (<1.98 vs. ≥1.98 mol%).

TABLE 3 Linear regression analyses of n3 LC-PUFAs and physical activity as predictors of individual cardiometabolic risk factors in midlife adults¹

Model ²	Variable	Mean BP			Waist circumference			HDL-C			Triglycerides			Glucose		
		R ²	β (95%CI)	P	R ²	β (95%CI)	P	R ²	β (95%CI)	P	R ²	β (95%CI)	P	R ²	β (95%CI)	P
1	EPA+DHA	0.21	-0.09 (-0.19, 0.01)	0.06	-0.05 (-0.14, 0.05)	0.32	0.22	0.06 (-0.04, 0.16)	0.23	0.10	-0.01 (-0.11, 0.10)	0.88	0.05	0.00 (-0.11, 0.11)	0.99	
	Physical activity			0.49	-0.16 (-0.25, -0.07)	0.001		0.13 (0.03, 0.23)	0.01		-0.14 (-0.24, -0.03)	0.01		-0.010 (-0.21, 0.01)	0.07	
2	Interaction	0.22	-0.09 (-0.19, 0.00)	0.06	-0.13 (-0.21, 0.03)	0.007	0.24	0.12 (0.03, 0.22)	0.01	0.12	-0.15 (-0.25, -0.05)	0.005	0.05	-0.07 (-0.17, 0.04)	0.23	
	AA/(EPA+DHA)	0.21	0.08 (-0.02, 0.18)	0.10	0.02 (-0.07, 0.11)	0.67	0.22	-0.05 (-0.15, 0.05)	0.31	0.10	0.00 (-0.10, 0.11)	0.99	0.07	0.04 (-0.07, 0.14)	0.50	
2	Physical activity			0.46	-0.17 (-0.26, -0.07)	<0.001		0.13 (0.03, 0.23)	0.009		-0.14 (-0.24, -0.03)	0.01		-0.09 (-0.20, 0.02)	0.09	
	Interaction	0.21	0.07 (-0.03, 0.16)	0.19	0.18 (0.08, 0.26)	<0.001	0.23	-0.09 (-0.19, 0.00)	0.05	0.12	0.14 (0.04, 0.24)	0.008	0.08	0.12 (0.02, 0.23)	0.02	

¹ AA, arachidonic acid; BP, blood pressure; HDL-C, HDL cholesterol; LC-PUFAs, long-chain, polyunsaturated fatty acids.

² Covariates are age, race, and sex in all models.

foster fat catabolism through PPAR activation may protect against such ectopic lipid deposition. Nonetheless, to date clinical trials have found no effect of fish oil supplementation on glycemic control in diabetics (6), and most trials examining effects of supplementation in nondiabetics have found no effect on insulin resistance (5).

More surprisingly, some analyses of longitudinally followed cohorts have found greater fish intake to be associated with a higher incidence of diabetes (7,9), and a recent cross-over study in diabetics reported higher glucose concentrations with a high n3 diet than with a high n6 diet (8). The latter experiment showed further that the n3 LC-PUFA diet lowered insulin concentrations throughout the day while insulin sensitivity during euglycemic clamp improved. This and other similar findings suggest that long-chain, n3 fatty acids may reduce release of insulin from β cells while enhancing peripheral insulin sensitivity (24,25). These multilevel effects on metabolism could give rise to clinical results that vary with other factors. For example, whereas no weight loss or metabolic benefit occurs in obese persons supplemented with n3 LC-PUFAs, when used in conjunction with an energy-restricted diet fish oil can enhance weight loss (26,27) and may magnify the fall in glucose and insulin concentrations achieved with energy restriction (28).

The current report examined the moderating effects of physical activity on associations between n3 LC-PUFAs, cardiometabolic risk, and insulin resistance. Aerobic exercise improves summary indices of cardiometabolic risk and each component risk factor (29) while also reducing visceral and liver fat stores (13). Daily physical activity measured by 6-d accelerometry correlates with insulin sensitivity determined by hyperinsulinemic, euglycemic clamp (30). The molecular processes within skeletal muscle affected by aerobic exercise include 1) increased glucose transport via GLUT4, 2) increased mitochondrial density and enzymatic activity, 3) increased vascularization, 4) improved endothelial function, and 5) healthy repartitioning of intracellular fat (29,31–33). The actions of EPA and DHA within myocytes may partially overlap with this list, but include complementary actions in hepatocytes and in adipose tissue (2).

The combination of exercise and fish consumption could produce uniquely beneficial metabolic effects. For example, sedentary lifestyle and obesity are both associated with chronic inflammation. Exercise acutely constitutes an oxidative stressor, yet habitual physical activity and fitness appear to reduce chronic markers of inflammation (34,35). This adaption to the acute, potentially deleterious effects of exercise could be facilitated by EPA and DHA through production of eicosanoids that are relatively anti-inflammatory or facilitate resolution of inflammation (36) or through the roles of EPA and DHA derivatives in quenching reactive oxygen species (1).

Excess adipose tissue leads to lipotoxic infiltration of muscle and solid organs, which exercise combats through increased oxidative capacity (37). By activating PPAR- α , EPA and DHA increase the expression of multiple genes regulating lipoprotein concentrations but particularly those involved with fatty acid oxidation. Thus, working in concert, habitual exercise and increased n3 LC-PUFA exposure may maximize utilization of fats as fuel by muscle tissue, averting triglyceride excess in serum, adipose tissue, and extra-adipose deposition.

Clinical experiments have found that the combination of aerobic exercise and fish oil supplementation results in a reduction in fat mass and postprandial lipemia not achieved by either intervention alone (38,39). Thus, the present findings may analogously reflect synergism between aerobic exercise and n3 LC-PUFAs across the various tissues involved in the regulation

of fuel metabolism. We have previously reported a similarly interactive effect of EPA and DHA exposure and physical activity on cardiac autonomic control (reflected in heart rate variability), whereby greater circulating levels of n3 LC-PUFAs were associated with greater heart rate variability only in persons expending >2000 kcal/wk in physical activity (40).

This investigation had certain strengths and limitations. The participants were community volunteers free of diabetes and any vascular disease. Also, based on additional medical exclusions and self-selection, the sample was healthier than our parent sample in several respects. Nonetheless, the participants in this substudy did not exercise more than excluded persons, and the presented analyses are free from the confounding effects of prescription drugs and supplements. The availability of serum phospholipid fatty acid composition permitted unbiased estimates of EPA and DHA exposure. Cardiometabolic risk was calculated as a continuous measure because this measure outperforms binary expressions in predicting cardiovascular disease events (20). The study's cross-sectional design precluded causal inference. Also, the study relied on HOMA-IR and self-reported physical activity, measures with limited fidelity compared with gold standard indicators of insulin sensitivity and exercise-related energy expenditure, respectively. Finally, the results were derived from a self-selected sample of healthy and largely Caucasian adult volunteers under 55 y of age; therefore, they may or may not hold for other populations.

Overall, the present findings expand on previous reports by newly suggesting that in midlife adults the cardiometabolic effects of n3 LC-PUFA exposure varies with concomitant physical activity, that is, beneficial effects of EPA and DHA accrue in physically active, but not in sedentary, individuals. This observation may underlie the discordant research to date on the effects of these fatty acids on diabetes and cardiometabolic risk. Because the molecular metabolic effects of n3 LC-PUFAs and exercise partially overlap and may be complementary, the current results are mechanistically plausible and testable in clinical trials and are potentially important to the prevention of diabetes and cardiovascular disease.

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