Research Article

Cancer Prevention Research

Association between Serum Phospholipid Fatty Acids and Intraprostatic Inflammation in the Placebo Arm of the Prostate Cancer Prevention Trial

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Abstract

Inflammation may play an etiologic role in prostate cancer. Several dietary factors influence inflammation; studies have shown that long-chain n-3 polyunsaturated fatty acids are anti-inflammatory, whereas n-6 and *trans* fatty acids are proinflammatory. We evaluated whether serum phospholipid n-3, n-6, and *trans* fatty acids were associated with intraprostatic inflammation, separately in 191 prostate cancer cases and 247 controls from the placebo arm of the Prostate Cancer Prevention Trial (PCPT). Men without a prostate cancer diagnosis underwent prostate biopsy at trial end, and benign prostate tissue inflammation was evaluated in approximately three biopsy cores per man; this was expressed as no, some, or all cores with inflammation. In controls, serum eicosapentaenoic acid [OR of all cores with inflammation versus none (95% CI), 0.35 (0.14–0.89)] and docosahexaenoic acid [OR (95% CI), 0.42 (0.17–1.02)] were inversely associated with, whereas linoleic acid [OR (95% CI), 3.85 (1.41–10.55)] was positively associated with intraprostatic inflammation. Serum *trans* fatty acids were not associated with intraprostatic inflammation. No significant associations were observed in cases; however, we could not rule out a positive association with linoleic acid and an inverse association with arachidonic acid. Thus, in the PCPT, we found that serum n-3 fatty acids were inversely, n-6 fatty acids were positively, and *trans* fatty acids were not associated with intraprostatic inflammation in controls. Although, in theory, inflammation could mediate associations of serum fatty acids with prostate cancer risk, our findings cannot explain the epidemiologic associations observed with n-3 and n-6 fatty acids. *Cancer Prev Res; 8(7); 590–6. ©2015 AACR.*

Introduction

Inflammation may play an etiologic role in prostate carcinogenesis (1, 2). Supporting evidence indicates that several single nucleotide polymorphisms (SNP) in inflammation-related pathways are associated with prostate cancer risk (2), and use of nonsteroidal anti-inflammatory drugs is inversely associated with prostate cancer risk (3). Several dietary factors have been shown to influence inflammation in both human (1, 4, 5) and animal models (6, 7). In particular, long-chain n-3 polyunsaturated fatty acids (PUFA) been shown to be anti-inflammatory (2, 8), whereas both n-6 and *trans* fatty acids have been shown to be proinflammatory (9). However, to our knowledge, no studies have directly examined whether serum fatty acids are associated with intraprostatic inflammation, and thus could mediate previously observed inverse associations of serum n-3 fatty acid concentrations, or direct associations of serum n-6 fatty acid concentrations, with prostate cancer risk (10, 11).

To address this question, we examined associations of inflammation-related serum phospholipid fatty acids with the prevalence and extent of inflammation in benign prostate tissue, in a subset of prostate cancer cases and controls randomized to the placebo arm of the SWOG-coordinated Prostate Cancer Prevention Trial (PCPT). Using data from these same men, Gurel and colleagues recently reported that both the prevalence and extent of intraprostatic inflammation were positively associated with prostate cancer; the strongest associations were observed with high-grade cancers (12). With the goal of improving our understanding of prostate cancer etiology and providing information that may inform dietary prevention strategies to reduce prostate cancer risk, we conducted a cross-sectional study in which we

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Note: Supplementary data for this article are available at Cancer Prevention Research Online (http://cancerprevres.aacrjournals.org/).

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hypothesized that serum n-3 fatty acids would be inversely associated with intraprostatic inflammation, and that serum n-6 and *trans* fatty acids would be positively associated with intraprostatic inflammation.

Materials and Methods

Study design and population

The PCPT was a multicenter, randomized, placebo-controlled trial testing whether the 5α-reductase type II inhibitor, finasteride, reduced prostate cancer risk (13). Between January 1993 and May 1997, and across 221 study centers in the United States, 18,882 men were randomized to finasteride (5 mg/day) or placebo for 7 years. Eligible men were at least 55 years old, had a prostate-specific antigen (PSA) level ≤3.0 ng/mL, a normal digital-rectal examination (DRE), and no history of cancer (except nonmelanoma skin) or severe lower urinary tract symptoms. At trial entry, men completed self-administered questionnaires on demographic, lifestyle, and medical factors, including age, race, alcohol consumption, smoking history, diabetes status, physical activity, and family history of prostate cancer (14, 15). In addition, weight and height were measured, and body mass index (BMI) calculated [weight (kg)/height $(m)^2$].

Men underwent annual PSA and DRE; those with abnormal DRE or PSA level ≥4.0 ng/mL were recommended for prostate biopsy (13). Cancers detected during these biopsies were considered to be "for-cause" biopsy detected. All men who had not been diagnosed with prostate cancer during the trial were requested to undergo a prostate biopsy at the end of the trial, regardless of PSA level or DRE result. Cancers detected on an end of study biopsy were considered to be "for-cause" biopsy detected if there was an indication (i.e., abnormal PSA or DRE), or "not-for-cause" biopsy detected if there was no clinical indication for biopsy.

Prostate cancer diagnosis made at the study site was confirmed, and Gleason sum determined, centrally at the Prostate Diagnostic Laboratory, University of Colorado, Colorado; pathologists were blinded to trial arm and exposure information. The Data and Safety and Monitoring Committee stopped the PCPT early, because men receiving finasteride showed a statistically significant 25% lower period prevalence of prostate cancer than men receiving placebo (13). The Institutional Review Boards at individual participating trial sites approved the PCPT. The Institutional Review Boards at the Johns Hopkins Bloomberg School of Public Health and the Colorado Multiple Institutional Review Board approved this study.

Selection of cases and controls

For the prior study on inflammation and prostate cancer (12), cases ($N = 191$) and controls ($N = 209$) were selected from the PCPT nested case–control study (16). Controls were frequency matched to cases on age at baseline, family history of prostate cancer, and treatment arm; they were also oversampled for nonwhite men. For this study, this was supplemented with 38 controls from the placebo arm for a total of 247 controls. These additional controls had no clinical indication for biopsy during all 7 years of the trial or at the time of the end of study biopsy to allow for subanalyses restricted to controls without indication; these controls were also frequency matched to the cases on age and family history. For these additional controls, an approximately equal number of cases by grade (Gleason sum: <7 or 7–10), and by reason for biopsy (for cause, not for cause) were selected.

Assessment of inflammation in benign biopsy tissue

The assessment of inflammation in benign tissue from biopsy cores for the 191 cases and 247 controls has been previously described (12). Briefly, the prevalence and extent of inflammation in benign prostate tissue was assessed on H&E-stained slides that were used to make or exclude the diagnosis of prostate cancer. A mean of 3.3 biopsy cores, primarily from the apex or mid-gland, per man were reviewed for inflammation by a single pathologist (B. Gurel). To ensure blinding to case–control status, all areas of adenocarcinoma (cases) and arbitrary benign areas on cores without cancer (cases and controls) were masked with ink on the slide cover slips. The H&E-stained slides were then digitized using the Aperio ScanScope slide scanner (Aperio) and uploaded into the Spectrum Digital Pathology Information Management System (Aperio). Slide images were reviewed for inflammation using Aperio ImageScope Viewer. For this study, inflammation data were expressed as prevalence and extent, as previously described (12). Prevalence of inflammation was defined as the presence of any inflammatory cells, either acute or chronic, in the benign tissue of any core assessed. Extent of inflammation was defined as none, some, or all biopsy cores containing any inflammatory cells.

Assessment of serum fatty acids

Approximately 15 mL of nonfasting blood was collected from each participant 3 months prior to randomization, and annually thereafter until diagnosis or end of study. Venous blood was drawn into collection tubes without anticoagulant and stored at room temperature for 30 to 60 minutes before centrifugation. The serum fraction was then separated and frozen as quickly as possible before being shipped to the specimen repository, where the samples were stored at −70°C until analysis (14, 17). Serum samples were collected at 1 and 4 years after randomization, and 0.5 mL aliquots were pooled before analysis to reduce intraindividual variability. Alternate years were selected if men were missing a year 1 or year 4 sample or were diagnosed before year 4 ($n = 69$), and a single, prediagnostic sample was used if 2 prediagnostic blood samples were unavailable ($n = 1$). The average (SD) time between the most recent blood draw used for analysis and time of biopsy was 189 days (228) for controls, and 176 days (187) for cases.

Detailed methods for the phospholipid fatty acid assay have been published elsewhere (18, 19). Briefly, total lipids were extracted from serum, and phospholipids were separated from other lipids by one-dimensional thin-layer chromatography (20). Methyl esters of phospholipid fatty acids were prepared by direct transesterification (21) and separated by gas chromatography using Supelco-fused silica 100-m capillary column SP-2560. Fatty acid composition was expressed as the weight percentage of total phospholipid fatty acids. Pooled quality control samples were embedded randomly in each box of study samples. Samples from cases and controls were analyzed simultaneously, and all laboratory personnel were blinded to the case–control status of the samples. Coefficients of variation for fatty acids of the quality control samples were as follows: 18:3 n-3, 5.1%; 20:4 n-6, 1.0%; 22:6 n-3, 2.4%; 20:5 n-3, 3.0%; 18:2 n-6, 1.5%; TFA 16, 10.1%; TFA 18:1, 7.3%; and TFA 18:2, 10.3%. There was no evidence of laboratory drift.

Statistical analyses

Associations of serum fatty acids and intraprostatic inflammation were analyzed separately in cases and controls. Due to

nonnormal distributions, serum fatty acids were natural logarithm transformed for calculation of geometric means. Serum fatty acids were categorized into tertiles for both cases and controls on the basis of their distributions in inflammation-free controls; results for cases were not qualitatively different when we used case-specific tertile cutpoints. The following fatty acid variables were calculated: the sum of eicosapentaenoic acid (EPA; 20:5 n-3), docosahexaenoic acid (DHA; 22:6 n-3), and docosapentaenoic acid (DPA; 22:5 n-3) as a measure of total long chain n-3 fatty acids; the sum of linoleic (18:2 n-6) and arachidonic acids (20:4 n-6) as a measure of total n-6 fatty acids; the sum of 18:1 n-6t, 18:1 n-7t, 18:1 n-8t, and 18:1 n-9t as a measure of total TFA 18:1; the sum of trans-fats16:1 n-7t and 16:1 n-9t as a measure of total TFA 16:1; and the sum of trans-fats 18:2 n-6tt, 18:2 n-6ct, and 18:2 n-6tc as a measure of total TFA 18:2. These summed variables were then natural logarithm transformed for calculation of geometric means.

Least square mean values of demographic characteristics, stratified by prevalence and extent of inflammation, were adjusted for age, family history of prostate cancer, and race using linear or logistic regression models for continuous and categorical variables, respectively. Tests for linear trend (P_{trend}) across inflammation categories were based on linear or logistic regression models using an ordinal variable to describe extent of inflammation from lowest (no cores with inflammation) to highest (all cores with inflammation).

Differences in serum fatty acid levels by inflammation status were calculated using linear regression models. Geometric mean serum fatty acid concentrations were adjusted for age, family history of prostate cancer, and race using the residual method (22). Tests for linear trend (P_{trend}) in fatty acid concentrations across inflammation categories were based on logistic regression models using an ordinal variable to describe extent of inflammation.

Multivariable-adjusted multinomial logistic regression models were used to estimate odds ratios (OR) and 95% confidence intervals (CI) for the associations of fatty acid concentrations with extent of inflammation. Models were adjusted for age, race, and family history of prostate cancer. In these models, tests for linear trend (P_{trend}) were based on multinomial logistic regression models evaluating associations of extent of inflammation with log-transformed serum fatty acid concentrations modeled as a continuous variable. Analyses were repeated in the subset of 223 controls with a "not-for-cause" biopsy. All statistical analyses were performed using Stata I/C, version 13 (StataCorps). All tests were two-sided and $P < 0.05$ was considered to be statistically significant.

Results

Table 1 gives baseline demographic and lifestyle characteristics for controls with and without intraprostatic inflammation. Compared to controls with no inflammation, controls with inflammation were more likely to be older, and have a higher PSA concentration at baseline. There were no other differences between men with or without inflammation in controls.

Table 2 gives geometric means and 95% CIs of serum n-3, n-6, and *trans* fatty acid concentrations by extent of inflammation in controls. Serum concentrations of EPA, DHA, total n-3, linoleic acid, and total n-6 fatty acids, but not α-linolenic acid, arachidonic acid, or any of the *trans* fatty acids, differed by extent of intraprostatic inflammation. Specifically, mean serum EPA, DHA, and total n-3 fatty acids concentrations decreased, whereas linoleic acid and total n-6 fatty acids increased with extent of inflammation.

Table 3 gives adjusted ORs for associations of serum fatty acids with extent of intraprostatic inflammation in controls. In these men, serum levels of EPA, DHA, and total n-3 fatty acids were significantly inversely associated with the extent of inflammation; the odds of having all cores with inflammation (versus none) was 65%, 58%, and 65% lower in those in the highest tertile of serum EPA, DHA, and total n-3 fatty acids, respectively, relative to those in the lowest tertile. In contrast, serum linoleic and total n-6 fatty acids were positively associated with the extent of inflammation; the odds of having all cores with inflammation was 3.85 and 3.95 times higher in those in the highest tertile of serum linoleic and total n-6 fatty acids, respectively, relative to those in the lowest tertile. None of the *trans* fatty acids was associated with intraprostatic inflammation in controls. Results were not qualitatively

Table 1. Characteristics[a] of controls with and without inflammation in prostate biopsy cores, in the placebo arm of the PCPT

	At least one biopsy core with inflammation			Extent of biopsy cores with inflammation		
	No	Yes	*P*	Some	All	P_{trend}
n	55	192		127	65	
Mean age at baseline (years)	62.6	64.6	0.021	64.3	65.1	0.015
Mean age at biopsy (years)	71.1	71.1	0.70	71.1	71.1	0.98
Non-white (%)	14.5	17.2	0.64	17.3	16.9	0.74
Family history (%)	16.4	18.2	0.75	17.2	20.0	0.60
Cigarette smoking history (%)						
Current or former	71.8	66.4	0.55	65.2	68.8	0.72
Never	28.2	33.6		34.8	31.2	
Mean pack-years smoked, current and former smokers	15.9	17.1	0.91	16.3	18.7	0.57
Mean body mass index (kg/m^2)	27.8	27.4	0.56	27.3	27.7	0.92
Moderately active or active (%)	52.7	47.7	0.23	53.1	36.9	0.091
History of diabetes (%)	12.9	7.8	0.25	8.6	6.1	0.20
Mean PSA						
Concentration at baseline (ng/mL)	0.9	1.2	0.001	1.2	1.3	0.001
Concentration at biopsy (ng/mL)	1.3	2.2	0.25	1.5	3.4	0.019
Velocity (ng/mL/year)	0.06	0.11	0.43	0.05	0.21	0.03

[a]Means are least squares means for all characteristics except age at baseline, family history of prostate cancer, and race. Least squares means and *P*-values were estimated using linear regression models for continuous characteristics or logistic regression models for binary characteristics, and were adjusted for baseline age, family history, and race.

Table 2. Distribution of serum phospholipid fatty acids among controls with and without inflammation in prostate biopsy cores, in the placebo arm of the PCPT

	No biopsy cores with inflammation		Some biopsy cores with inflammation		All biopsy cores with inflammation		
Fatty acid	Mean[a,b]	95% CI	Mean[a]	95% CI	Mean[a]	95% CI	P_{trend}
n-3 fatty acids							
Total n-3 fatty acids	4.30	4.02–4.59	4.37	4.19–4.56	3.80	3.58–4.04	0.004
α-Linolenic acid	0.13	0.12–0.14	0.14	0.13–0.15	0.14	0.13–0.15	0.35
EPA	0.59	0.53–0.67	0.59	0.54–0.63	0.49	0.44–0.55	0.016
DHA	2.78	2.58–3.00	2.87	2.73–3.01	2.46	2.30–2.64	0.012
DPA	0.87	0.82–0.92	0.85	0.82–0.88	0.80	0.76–0.85	0.037
n-6 fatty acids							
Total n-6 fatty acids	30.85	30.35–31.36	30.98	30.65–31.31	31.73	31.26–32.20	0.010
Linoleic acid	19.36	18.73–20.01	19.73	19.31–20.16	20.40	19.78–21.03	0.022
Arachidonic acid	11.21	10.70–11.74	11.01	10.68–11.35	11.06	10.60–11.55	0.70
Trans fatty acids							
TFA 18:1	1.07	0.98–1.18	1.01	0.95–1.07	1.07	0.99–1.18	0.90
TFA 18:2	0.22	0.21–0.24	0.21	0.20–0.22	0.22	0.21–0.23	0.77
TFA 16:1	0.23	0.21–0.24	0.23	0.22–0.24	0.23	0.22–0.24	0.68

Abbreviations: DHA, docosahexaenoic acid; DPA, docosapentaenoic acid; EPA, eicosapentaenoic acid; TFA, *Trans* fatty acids.

[a]Serum fatty acids are presented as percent of total serum phospholipid fatty acids.

[b]Geometric least squares means and *P*-values were estimated using linear regression models, adjusting for baseline age, family history, and race.

different when restricting analyses to controls with a not-for-cause biopsy ($n = 223$; data not shown).

Baseline demographic and lifestyle characteristics for men with prostate cancer are given in Supplementary Table S1. Compared to cases with no inflammation, cases with inflammation were more likely to be older, and non-white. Supplementary Table S2 gives geometric means and 95% CIs of serum n-3, n-6, and *trans* fatty acids concentrations, by extent of inflammation for cases; mean serum fatty acid levels did not differ by the extent of inflammation in these men. There were no statistically significant associations of serum fatty acids and extent of intraprostatic inflammation in cases; these data are presented in Supplementary Table S3. However, we could not rule out a possible positive association for linoleic acid and inverse association for arachidonic acid. Results were not qualitatively different when restricting to cases with a for-cause ($n = 94$) or not-for-cause ($n = 97$) biopsy (data not shown).

Discussion

In the PCPT, we found that serum concentrations of anti-inflammatory n-3 fatty acids were inversely, whereas serum concentrations of proinflammatory n-6 fatty acids were positively associated with the extent of intraprostatic inflammation in controls. *Trans* fatty acids were not associated with the extent of intraprostatic inflammation in controls. In men with prostate cancer, there were no statistically significant associations of serum phospholipid fatty acids and intraprostatic inflammation; however, we could not rule out a possible positive association for linoleic acid and inverse association for arachidonic acid.

Given the direct association of intraprostatic inflammation with total and high-grade prostate cancer risk (12), our finding that serum n-3 fatty acids were inversely associated with intraprostatic inflammation in men without a diagnosis of prostate cancer is compatible with studies that have observed inverse associations of n-3 fatty acids and prostate cancer risk. This includes findings from both the Physician's Health study and the NIH-AARP study (10, 11). However, our findings cannot explain the positive association of DHA, an n-3 fatty acid, with high-grade prostate cancer risk recently observed in the PCPT (18), SELECT (23), and EPIC (24) studies, or the positive associations of EPA and DPA with total prostate cancer risk observed in a recent pooled analysis (25). It is possible that these counterintuitive findings are explained by residual confounding by measured or unmeasured healthy lifestyle factors, including the uptake of PSA-based prostate cancer screening (26). Alternatively, associations of n-3 serum fatty acids with prostate cancer risk may not be mediated through an inflammatory pathway.

In the PCPT, serum *trans* fatty acids were inversely associated with prostate cancer risk, whereas serum n-6 fatty acids were not associated with risk (18). The findings of this study suggest that the former association is not mediated by prostatic inflammation as we did not observe an association between *trans* fatty acids and intraprostatic inflammation in men without prostate cancer. Our results suggesting that n-6 fatty acids are positively associated with intraprostatic inflammation are not compatible with the null finding for n-6 fatty acids and prostate cancer risk in the PCPT. A recent study in the PCPT observed that inflammation in benign prostate tissue was positively associated with odds of prostate cancer, particularly high-grade disease, in the same subset of PCPT participants studied here (12). That there was no association between n-6 fatty acids and prostate cancer risk observed in the larger nested case–control sample of PCPT participants and in other studies (24, 27, 28) suggests that associations of n-6 fatty acids with prostate cancer risk are also not mediated through an inflammatory pathway.

We found no statistically significant associations between serum fatty acids and intraprostatic inflammation in prostate cancer cases. However, we could not rule out possible associations for linoleic acid and arachidonic acid; these nonsignificant associations were of similar magnitude and direction as observed in controls. We cannot rule out that inflammation in benign prostate tissue measured in men with a diagnosis of prostate cancer may have occurred as a response to the presence of the cancer.

This study has several strengths and limitations that warrant discussion. The primary strength of this study was that we were able to assess the fatty acid–tissue inflammation association in men without a diagnosis of prostate cancer. Most of these men did not have a clinical indication for biopsy and all were biopsied at the end of study biopsy. Thus, we were able to examine associations between serum fatty acids and inflammation in the prostate directly, rather than evaluating associations with inflammation

Table 3. Multivariable-adjusted associations of serum phospholipid fatty acids in relation to inflammation in prostate biopsy cores, among controls from the placebo arm of the PCPT[a]

	No biopsy cores with inflammation	Some biopsy cores with inflammation		All biopsy cores with inflammation	
Fatty acid	***n***	***n***	**OR (95% CI)**	***n***	**OR (95% CI)**
n-3 fatty acids					
Total n-3 fatty acids					
T1	18	37	1.00 (ref)	30	1.00 (ref)
T2	18	52	1.44 (0.66–3.18)	23	0.62 (0.26–1.49)
T3	19	38	1.02 (0.46–2.27)	12	0.32 (0.13–0.83)
P-trend			0.57		0.007
α-Linolenic acid (18:3 n3)					
T1[a]	19	43	1.00 (ref)	21	1.00 (ref)
T2	18	26	0.63 (0.27–1.43)	11	0.53 (0.19–1.43)
T3	18	58	0.63 (0.66–3.04)	33	1.64 (0.69–3.87)
P_{trend}			0.25		0.32
EPA (20:5 n3)					
T1	20	57	1.00 (ref)	37	1.00 (ref)
T2	17	35	0.65 (0.29–1.43)	17	0.46 (0.19–1.12)
T3	18	35	0.71 (0.33–1.54)	11	0.35 (0.14–0.89)
P_{trend}			0.90		0.017
DHA (22:6 n3)					
T1	18	40	1.00 (ref)	33	1.00 (ref)
T2	18	37	0.92 (0.41–2.04)	16	0.48 (0.19–1.18)
T3	19	50	1.11 (0.51–2.41)	16	0.42 (0.17–1.02)
P_{trend}			0.50		0.017
DPA (22:5 n-3)					
T1	19	45	1.00 (ref)	27	1.00 (ref)
T2	17	48	1.19 (0.54–2.61)	21	0.84 (0.35–2.05)
T3	19	34	0.75 (0.34–1.64)	17	0.60 (0.25–1.48)
P_{trend}			0.44		0.28
n-6 fatty acids					
Total n-6 fatty acids					
T1	19	33	1.00 (ref)	10	1.00 (ref)
T2	17	47	1.52 (0.68–3.37)	16	1.63 (0.57–4.63)
T3	19	47	1.43 (0.65–3.12)	39	3.95 (1.52–10.29)
P_{trend}			0.69		0.011
Linoleic acid (18:2 n-6)					
T1	18	31	1.00 (ref)	9	1.00 (ref)
T2	19	53	1.70 (0.76–3.78)	24	2.66 (0.96–7.40)
T3	18	43	1.46 (0.65–3.29)	32	3.85 (1.41–10.55)
P_{trend}			0.35		0.024
Arachidonic acid (20:4 n-6)					
T1	18	50	1.00 (ref)	22	1.00 (ref)
T2	18	32	0.66 (0.29–1.48)	22	1.03 (0.42–2.55)
T3	19	45	0.83 (0.38–1.79)	21	0.87 (0.35–2.13)
P_{trend}			0.52		0.67
Trans fatty acids					
TFA 18:1					
T1	9	29	1.00 (ref)	6	1.00 (ref)
T2	9	24	0.84 (0.28–2.56)	12	2.01 (0.50–8.08)
T3	38	74	0.58 (0.24–1.37)	47	1.83 (0.58–5.71)
P_{trend}			0.57		0.61
TFA 18:2					
T1	19	54	1.00 (ref)	22	1.00 (ref)
T2	18	37	0.66 (0.30–1.44)	25	1.05 (0.43–2.53)
T3	18	36	0.65 (0.30–1.43)	18	0.78 (0.31–1.95)
P_{trend}			0.24		0.72
TFA 16:1					
T1	18	36	1.00 (ref)	20	1.00 (ref)
T2	18	53	1.42 (0.65–3.12)	22	1.04 (0.42–2.58)
T3	19	38	0.93 (0.42–2.08)	23	1.00 (0.41–2.45)
P_{trend}			0.66		0.67

Abbreviations: DHA, docosahexaenoic acid; DPA, docosapentaenoic acid; EPA, eicosapentaenoic acid; TFA, *Trans* fatty acids.

[a]OR calculated using polytomous logistic regression, where "no biopsy cores with inflammation" served as the outcome reference category, and fatty acid tertile 1 served as the exposure reference category. OR were adjusted for age, race, and family history of prostate cancer.

biomarkers or other surrogates of prostatic inflammation. In addition, measurement error due to intra-individual variability in serum fatty acid measures was reduced in this study through the use of two pooled blood draws for analysis; however, it should also be noted that serum phospholipid fatty acid measures reflect short-term dietary intake, relative to whole blood measures.

The primary limitation of this study was its small sample size; this restricted our statistical power and limited the ability to directly assess whether the associations of serum fatty acids with prostate cancer risk were or were not mediated by intraprostatic inflammation. Furthermore, inflammation was expressed as the prevalence and extent of inflammation irrespective of inflammatory cell type. Future studies should consider associations with specific inflammatory cell types. Although we performed multiple statistical tests, this study is the first, to our knowledge, to investigate serum fatty acids with tissue inflammation. Thus, we chose not to adjust the alpha level for number of tests performed. In addition, the associations presented here may, in part, be explained by confounding or residual confounding (26). Serum fatty acids were measured as a proportion of total serum fatty acids. When expressed in this way, a positive association with one fatty acid could result in a false inverse association with another; we did not attempt to isolate the independent associations of each fatty acid in the present analysis due to a lack of statistical power. Finally, although the men we studied were from the placebo arm of a chemoprevention trial and were annually screened for prostate cancer, we have no reason to believe that the associations we observed between serum fatty acid concentrations and intraprostatic inflammation would be different from the general population of similarly aged U.S. men.

In conclusion, we found that serum n-3 fatty acids were associated with decreased odds of inflammation, and serum n-6 fatty acids were associated with increased odds of inflammation, in benign prostatic tissue from men without prostate cancer in the placebo arm of the PCPT. Given that intraprostatic inflammation was positively associated with prostate cancer risk in the PCPT (12), our results cannot explain the unexpected associations between fatty acids and prostate cancer risk previously reported in the PCPT and elsewhere. Therefore, it is unlikely that inflammation mediates associations of serum fatty acids and prostate cancer risk.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

Disclaimer

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