Serum Insulin, Glucose, Indices of Insulin Resistance, and Risk of Prostate Cancer

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Background The mitogenic and growth-stimulatory effects of insulin-like growth factors appear to play a role in prostate carcinogenesis, yet any direct association of circulating insulin levels and risk of prostate cancer remains unclear.

Methods We investigated the relationship of the level of serum insulin, glucose, and surrogate indices of insulin resistance (ie, the molar ratio of insulin to glucose and the homeostasis model assessment of insulin resistance [HOMA-IR]) to the development of prostate cancer in a case–cohort study within the Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study cohort of Finnish men. We studied 100 case subjects with incident prostate cancer and 400 noncase subjects without prostate cancer from the larger cohort. Fasting serum was collected 5–12 years before diagnosis. We determined insulin concentrations with a double-antibody immunochemiluminometric assay and glucose concentrations with a hexokinase assay. Multivariable logistic regression models estimated relative risks as odds ratios (ORs), and all statistical tests were two-sided.

Results Insulin concentrations in fasting serum that was collected on average 9.2 years before diagnosis among case subjects were 8% higher than among noncase subjects, and the molar ratio of insulin to glucose and HOMA-IR were 10% and 6% higher, respectively, but these differences were not statistically significant. Among subjects in the second through fourth insulin quartiles, compared with those in the first quartile, increased insulin levels were associated with statistically significantly increased risks of prostate cancer (OR = 1.50, 95% confidence interval [CI] = 0.75 to 3.03; OR = 1.75, 95% CI = 0.86 to 3.56; and OR = 2.55, 95% CI = 1.18 to 5.51; for the second through fourth insulin quartiles, respectively; P_{trend} = .02). A similar pattern was observed with the HOMA-IR (OR = 2.10, 95% CI = 1.03 to 4.26; P_{trend} = .02) for the highest vs lowest quartiles. Risk varied inconsistently with glucose concentration (P_{trend} = .38). A stronger association between insulin level and prostate cancer risk was observed among leaner men and among men who were less physically active at work. Crude prostate cancer incidence was 154 prostate cancers per 100 000 person-years in the lowest quartile of fasting serum insulin vs 394 prostate cancers per 100 000 person-years in the highest quartile.

Conclusion Elevated fasting levels of serum insulin (but not glucose) within the normal range appear to be associated with a higher risk of prostate cancer.

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Prostate cancer is the most common malignancy in men in the United States, Canada, and Australia, and the prevalence of prostate cancer is rising elsewhere. The incidence of prostate cancer increases sharply with age and is higher among African American men and those with a family history of the disease in a first-degree relative, but little else is known about its etiology. Circulating androgens have long been thought to play a role in this hormone-dependent neoplasm, and growth factors, such as insulin-like growth factor I, have been found to be associated with the risk of prostate cancer, although evidence for strong or convincing causal relationships are lacking or inconsistent, including recent results from large pooled analyses (1–3). Most recently, genome-wide scans have revealed a number of chromosomal loci whose expression were modestly associated with risk of prostate cancer (4).

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The peptide hormone insulin is integrally involved in the homeostatic regulation of glucose and energy metabolism. In response to elevated levels of circulating glucose, insulin is rapidly secreted into the systemic circulation by pancreatic islet beta cells, after which it can activate cell-membrane insulin receptors and lead to increased uptake of glucose, proteins, and other molecules and an anabolic state. Insulin also has potent mitogenic and growth-stimulatory effects on the prostate and other tissues, and alterations in these effects could potentially contribute to the development of malignancy (5). The latter is supported by research linking insulin resistance to cancer risk (6), whereas research to date regarding a direct causal role for hyperinsulinemia in prostate cancer is limited and inconsistent, with three cohort studies (7–9) finding no association between insulin and risk, a case–control study (10) in China finding a strong positive relationship, and a recent report (11) from Sweden finding an inverse association between the proinsulin cleavage product, C-peptide, and the prostate cancer risk. Possible factors contributing to these study differences include retrospective vs prospective designs, length of follow-up, characteristics of the sample population (eg, race, relative weight, and disease stage at diagnoses), and whether plasma or serum was obtained from fasting or nonfasting participants.

To investigate whether fasting serum insulin, glucose, or indices of insulin resistance prospectively influence the development of prostate cancer, we conducted a case–cohort analysis within a large male cohort. Special attention was paid to disease stage, time from blood collection to cancer diagnosis, and interactions with body mass index, physical activity, and dietary factors relevant to insulin resistance.

CONTEXT AND CAVEATS

Prior knowledge

Insulin-like growth factors have several effects, including mitogenic and growth-stimulatory effects, that may be involved in prostate carcinogenesis; however, the association of circulating insulin levels with prostate cancer risk is unclear.

Study design

A case–cohort study within the Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study cohort of Finnish men. Insulin and glucose concentrations were determined in fasting serum samples that were collected 5–12 years before diagnosis of prostate cancer.

Contribution

There may be an increasing risk of prostate cancer across the normal range of insulin concentrations, although no association was observed between overall risk of prostate cancer and serum glucose levels.

Implications

Additional investigations on the association between insulin level and risk of prostate cancer are warranted, especially in nonsmoking populations and other races and ethnicities.

Limitations

The number of case subjects with prostate cancer in this study was small. Subjects included only Finnish male smokers (all whites), and so the findings may not be generalizable to other populations.

From the Editors

Participants and Methods

Study Population

The Alpha-Tocopherol, Beta-Carotene Cancer Prevention (ATBC) Study was a randomized, double-blind, placebo-controlled, primary prevention trial that tested whether daily supplementation with α-tocopherol, β-carotene, or both could reduce the incidence of lung or other cancers among male smokers (12,13). The trial was registered as ClinicalTrials.gov number, NCT00342992. A total of 29133 men between the ages of 50 and 69 years who smoked at least five cigarettes per day were recruited from southwestern Finland between April 1, 1985, and June 30, 1988, and randomly assigned to one of four intervention groups on the basis of a 2 × 2 factorial design. Men were ineligible who had previously been diagnosed with cancer, who has another serious illness, or who reported current use of vitamin E (>20 mg/d), vitamin A (>20000 IU/d), or β-carotene (>6 mg/d). Participants received α-tocopherol (50 mg/d) as DL-α-tocopheryl acetate, β-carotene (20 mg/d) as *all-trans*-β-carotene, both supplements, or a placebo capsule daily for 5–8 years (median = 6.1 years). Follow-up of the ATBC Study cohort after the intervention continued through the Finnish Cancer Registry. This study was approved by the institutional review boards of the US National Cancer Institute and the National Public Health Institute of Finland. Written informed consent was obtained from each participant before randomization.

Selection of Case Subjects and Noncase Subjects

Case subjects were defined as subjects with incident prostate cancer (*International Classification of Diseases, Ninth Revision*, code 185) diagnosed at least 5 years after their baseline blood collection (range = 5–12 years); this approach was followed to minimize any effect of preclinical disease on serum biochemistry (14) and to permit examination of any interaction with long-term use of the trial supplements. To conserve nonrenewable serum samples among subjects known to have prostate cancer, 100 case subjects were randomly selected from the 507 case subjects who met these criteria. Medical records were reviewed centrally by two study oncologists to confirm the diagnoses and stage by American Joint Committee on Cancer criteria (15). Among the 100 case subjects, 98 had histology or cytology results available that were reviewed and confirmed by a pathologist. We randomly selected 400 noncase subjects from the entire ATBC Study cohort who were alive 5 years after their baseline blood collection and who were not diagnosed with prostate cancer to serve as a subcohort comparison group for other site-specific cancer studies of serum insulin, glucose, and insulin resistance (16,17).

Specimen and Data Collection

At baseline visits for the ATBC Study, a morning serum sample was collected after an overnight fast and stored at −70°C. A general risk factor and medical history questionnaire was completed at the same time that included information regarding smoking, physical activity, and self-reported illnesses; and height and weight were measured, with body mass index calculated as (weight in kilograms)/(height in meters)^2 (12). Participants also completed a validated food-frequency questionnaire at baseline that included

both portion size and frequency of consumption for 203 food items and 73 mixed dishes (18). This instrument was intended to measure usual consumption of various foods and beverages during the previous 12 months. Family history of prostate cancer was queried during follow-up in 1991–1992 and therefore was available for only 82 case subjects and 302 noncase subjects.

Laboratory Assays

Serum insulin concentrations were determined by use of a double-antibody immunochemiluminometric assay performed on an Access automated platform (Beckman Instruments, Chaska, MN). Glucose concentrations were determined on a Hitachi 912 Chemistry Analyzer that used a hexokinase (Boehringer Mannheim, Indianapolis, IN) reaction whose reaction product was measured by spectrophotometric absorption at 340 nm. Samples from case subjects and from noncase subjects and blinded quality control duplicate samples were included in each batch. The within-batch coefficient of variation for insulin was 3.5%, and the between-batch coefficient of variation was 3.6%. For glucose, these coefficients of variation were 1.1% and 2.2%, respectively. The homeostasis model assessment of insulin resistance [HOMA-IR, calculated as fasting insulin (expressed as microunits/milliliter) × fasting glucose (expressed as millimoles/liter)/22.5 (19)] and the molar ratio of insulin to glucose were calculated and analyzed as surrogate indices of insulin resistance.

Statistical Analysis

Baseline characteristics of case subjects and noncase subjects were compared by using χ^2 and Fisher exact tests (for categorical variables) and *t* tests (for continuous variables). Correlations were estimated by using Spearman rank-order coefficient. Unconditional logistic regression models were used to estimate the relative risk (RR) for prostate cancer that was associated with serum insulin, serum glucose, the insulin to glucose ratio, and HOMA-IR through odds ratios (ORs) and 95% confidence intervals (CIs). Cox proportional hazards models with participants weighted by the inverse of their sampling fraction were also conducted and gave similar, somewhat stronger results. Consequently, we have reported only findings from the logistic regression models. Quartiles of insulin and the other primary exposures were defined by the distribution among the noncase subjects and entered into the models as indicator variables, with the lowest quartile as the referent category. All multivariable models adjusted for baseline age and body mass index, with the stratified dietary models also including energy intake. Other potential prostate cancer risk factors including benign prostatic hyperplasia, physical activity, serum cholesterol, intervention group, height, vitamin and/or mineral supplement use, smoking years, and cigarettes per day did not confound the insulin or insulin resistance associations (ie, they were not statistically significantly associated with prostate cancer or they did not alter primary risk estimates by 10% or more) and were not entered into the final models. Tests for linear trend used scored categorical trend variables that assigned the quartile median to each person. Effect modification was assessed by including the cross-product term of the primary factor (eg, insulin) with the covariate of interest and by subgroup analyses defined as being less than or greater than or equal to the median value of continuous variables or discrete categorical characteristics (eg, occupation or intervention assignment). Crude prostate cancer incidence was calculated on the basis of the background incidence in the whole cohort. All statistical tests were two-sided, and analyses were performed with SAS software version 8.02 (SAS Institute, Inc, Cary, NC).

Results

Baseline Characteristics

The average (mean) time from blood collection at study entry to diagnosis of prostate cancer was 9.2 years, and the average follow-up time for the noncase subjects was 10.4 years. Twenty-eight percent of the case subjects were diagnosed in American Joint Committee on Cancer stage I, 41% in stage II, 15% in stage III, and 15% in stage IV. Compared with noncase subjects, the men who developed prostate cancer were, at baseline, older, longer term smokers, and less likely to have higher occupational activity than men who did not (Table 1). Positive histories of benign prostatic hyperplasia and prostate cancer were also more common in

Table 1. Baseline characteristics of prostate cancer case subjects and noncase subjects*

Characteristic	Case subjects (n = 100)	Noncase subjects (n = 400)	*P*
Age, y (SD)	59.0 (4.6)	56.4 (5.0)	<.001†
Height, cm (SD)	174.4 (5.9)	173.8 (6.0)	.38†
Weight, kg (SD)	80.8 (14.9)	80.5 (13.2)	.82†
BMI, kg/m^2 (SD)	26.5 (4.5)	26.6 (3.9)	.91†
No. of cigarettes per day (SD)	20.9 (9.0)	20.5 (8.4)	.66†
Years of smoking (SD)	37.5 (8.1)	34.8 (8.5)	<.005†
History of diabetes, %	4.0	4.5	1.0‡
History of BPH, %	6.0	2.5	.10‡
Family history of prostate cancer, %	7.6	4.3	.34‡
Occupational activity, % active	16	29	.01§
Vitamin supplement use, %	24	16	.07§
Energy intake, kcal/d (SD)	2832 (736)	2834 (822)	.97†
Dietary fat, g/d (SD)	106 (37)	106 (38)	.94†
Dietary carbohydrate, g/d (SD)	307 (90)	305 (96)	.84†
Dietary protein, g/d (SD)	106 (31)	104 (30)	.62†
Dietary cholesterol, mg/d (SD)	577 (256)	589 (271)	.68†
Serum cholesterol, mmol/L (SD)	6.33 (1.22)	6.34 (1.17)	.94†
Insulin, μU/mL (SD)	5.56 (3.29)	5.14 (4.08)	.28†
Glucose, mg/dL (SD)	105 (28)	104 (24)	.57†
Molar ratio of insulin to glucose (SD)	0.053 (0.027)	0.048 (0.031)	.18†
HOMA-IR (SD)	1.52 (1.21)	1.43 (1.81)	.58†

* BMI = body mass index; BPH = benign prostatic hyperplasia; HOMA-IR = homeostasis model assessment of insulin resistance (calculated as fasting insulin in microunits per milliliter × fasting glucose in millimoles per liter/22.5).

† The *t* test was used. All statistical tests were two-sided.

‡ Fisher exact test was used.

§ The χ^2 test was used.

case subjects but not statistically significantly so. Insulin concentrations in fasting serum at least 5 years before diagnosis were on average 8% higher in case subjects (5.56 μIU/mL) than in noncase subjects (5.14 μIU/mL; difference = 0.42 μIU/mL, 95% CI = −0.34 to 1.18 μIU/mL; $P_{difference}$ = .28), with the molar ratio of insulin to glucose and the HOMA-IR being 10% higher (0.053 vs 0.048, 95% CI = −0.001 to 0.01; P = .18) and 6% higher (1.52 vs 1.43, 95% CI = −0.21 to 0.38; P = .58), respectively, among the case subjects.

Among the subcohort of 400 noncase subjects, serum insulin was strongly correlated with HOMA-IR (r = .98, P < .001), the insulin to glucose ratio (r = .96, P < .001), body mass index (r = .59, P < .001), and serum glucose (r = .44, P < .001), and glucose was correlated with HOMA-IR and body mass index (r = .58 and .26, respectively, each P < .001). The insulin to glucose ratio and HOMA-IR were also related to body mass index (r = .55 and .57, respectively, each P < .001). Serum concentrations of insulin and glucose, the insulin to glucose ratio, and HOMA-IR were not statistically significantly associated with smoking level, energy or macronutrient intake, or physical activity. We also evaluated the relationship between fasting serum insulin levels and baseline characteristics by calculating age-adjusted mean values according to quartile of serum insulin within the noncase subjects in the subcohort (Table 2). Body weight, body mass index, history of diabetes, prostate cancer family history, serum glucose and cholesterol, the insulin to glucose ratio, and HOMA-IR increased across insulin quartiles; however, job-related activity decreased. Dietary factors were less clearly related to serum insulin levels.

Serum Concentrations of Insulin and Glucose, Molar Ratio of Insulin to Glucose, HOMA-IR, and Prostate Cancer Risk

In age- and body mass index–adjusted analyses, the association between fasting insulin concentration and risk of prostate cancer increased statistically significantly across insulin quartiles (OR = 1.50, 95% CI = 0.75 to 3.03; OR = 1.75, 95% CI = 0.86 to 3.56; and OR = 2.55, 95% CI = 1.18 to 5.51; for the second through fourth quartiles, respectively; P_{trend} = .02), as did the association between HOMA-IR and risk of prostate cancer (OR = 0.82, 95% CI = 0.40 to 1.65; OR = 1.17, 95% CI = 0.59 to 2.34; and OR = 2.10, 95% CI = 1.03 to 4.26; P_{trend} = .02) (Table 3). In contrast, the molar ratio of insulin to glucose and fasting glucose level were not statistically significantly associated with prostate cancer risk (eg, OR = 1.75, 95% CI = 0.83 to 3.67, for the fourth quartile of the insulin to glucose ratio, and OR = 1.43, 95% CI = 0.76 to 2.68, for the fourth quartile of serum glucose) (Table 3). Crude prostate cancer incidence was calculated to be 154 prostate cancers per 100 000 person-years in the lowest quartile of fasting serum insulin vs 394 prostate cancers per 100 000 person-years in the highest insulin quartile.

Exploratory subgroup analyses that were based on factors with potential relevance to the association between serum insulin concentration and prostate cancer risk found somewhat stronger risk associations (ie, in magnitude and P_{trend} values) for higher serum insulin among leaner men (OR = 3.89, 95% CI = 1.31 to 11.54; P_{trend} = .01); heavier smokers (OR = 3.79, 95% CI = 1.23 to 11.74; P_{trend} = .08); those with lower levels of occupational physical activity (OR = 3.66, 95% CI = 1.48 to 9.04; P_{trend} = .04); or those consuming fewer calories (OR = 4.09, 95% CI = 0.24 to 13.45; P_{trend} = .02), less protein (OR = 3.21, 95% CI = 0.99 to 10.42; P_{trend} = .04), less

Table 2. Age-adjusted mean baseline characteristics of the 400 noncase subjects in the subcohort by quartile of fasting serum insulin: Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study*

Characteristic	Quartile 1 (≤2.75 μU/mL)	Quartile 2 (>2.75 to ≤4.10 μU/mL)	Quartile 3 (>4.10 to ≤6.10 μU/mL)	Quartile 4 (>6.10 μU/mL)
Age, y	56.6	56.1	56.1	56.7
Height, cm	173.1	174.0	174.4	173.8
Weight, kg	72.5	75.9	82.5	91.2
BMI, kg/m^2	24.2	25.0	27.1	30.2
No. of cigarettes per day	20.6	20.7	19.4	21.3
No. of years of smoking	35.5	34.5	34.1	35.3
History of diabetes, %	1.0	1.0	3.9	12.5
History of BPH, %	2.9	1.1	2.1	3.9
Family history of prostate cancer, %	1.6	5.1	5.3	5.0
Occupational activity, % active	35.4	36.1	21.7	22.7
Vitamin supplement use, % yes	11.0	21.9	17.6	14.6
Energy intake, kcal/d	2862	2760	2781	2942
Dietary fat, g/d	104.2	102.3	105.3	113.4
Dietary carbohydrate, g/d	311.2	300.1	295.0	312.6
Dietary protein, g/d	101.5	100.1	104.7	108.8
Alcohol (ethanol) intake, g/d	24.0	19.5	16.8	18.1
Dietary cholesterol, mg/d	568	569	597	624
Serum cholesterol, mmol/L	6.18	6.22	6.56	6.40
Insulin, μU/mL	2.04	3.47	5.05	10.22
Glucose, mg/dL	94.3	99.5	105.3	115.7
Molar ratio of insulin to glucose	0.02	0.04	0.05	0.09
HOMA-IR	0.48	0.85	1.32	3.17

* BMI = body mass index; BPH = benign prostatic hyperplasia; HOMA-IR = homeostasis model assessment of insulin resistance (calculated as fasting insulin in microunits per milliliter × fasting glucose in millimoles per liter/22.5).

Table 3. Age- and body mass index–adjusted associations of quartiles of baseline serum insulin, glucose, molar ratio of insulin to glucose, and homeostasis model assessment of insulin resistance (HOMA-IR) with prostate cancer risk: Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study*

Serum concentration, ratio, or index	Case subjects (n = 100)	Noncase subjects† (n = 400)	OR (95% CI)
Insulin, μU/mL			
≤2.75	17	100	1.00 (ref)
>2.75 to ≤4.10	23	101	1.50 (0.75 to 3.03)
>4.10 to ≤6.10	27	103	1.75 (0.86 to 3.56)
>6.10	33	96	2.55 (1.18 to 5.51)
			P_{trend} = .02
Glucose, mg/dL			
≤93	24	113	1.00 (ref)
>93 to ≤99	28	100	1.33 (0.72 to 2.48)
>100 to ≤107	17	88	0.92 (0.46 to 1.86)
>107	31	99	1.43 (0.76 to 2.68)
			P_{trend} = .38
Molar ratio of insulin to glucose			
≤0.03	20	100	1.00 (ref)
>0.03 to ≤0.04	22	100	1.11 (0.56 to 2.19)
>0.04 to ≤0.06	27	100	1.41 (0.71 to 2.78)
>0.06	31	100	1.75 (0.83 to 3.67)
			P_{trend} = .12
HOMA-IR			
≤0.69	22	100	1.00 (ref)
>0.69 to ≤1.02	17	100	0.82 (0.40 to 1.65)
>1.02 to ≤1.53	23	100	1.17 (0.59 to 2.34)
>1.53	38	100	2.10 (1.03 to 4.26)
			P_{trend} = .02

* HOMA-IR: calculated as fasting insulin in microunits per milliliter × fasting glucose in millimoles per liter/22.5. CI = confidence interval; OR = odds ratio; ref = referent.

† Noncase subjects are not evenly distributed across quartiles of insulin and glucose because several participants had identical insulin or glucose values.

carbohydrate (OR = 6.20, 95% CI = 1.72 to 22.35; P_{trend} = .005), or more alcohol (OR = 5.70, 95% CI = 1.60 to 20.28; P_{trend} = .01) (Table 4). Only the insulin–body mass index and insulin–occupational activity interaction tests ($P_{interaction}$ = .06 and .04, respectively) approached or achieved statistical significance, however. Use of World Health Organization definitions for being overweight (body mass index of >25 kg/m^2) and for obesity (body mass index of >30 kg/m^2) did not materially alter the interaction with body mass index (data not shown). The protein and carbohydrate interactions were independent of total energy intake, and the latter was evident only for complex carbohydrates (ie, starch and fiber; data not shown). Higher insulin levels were associated primarily with the development of earlier stage disease (OR = 3.19, 95% CI = 1.25 to 8.13; P_{trend} = .02), but there was no statistically significant effect modification based on the two periods of time from baseline blood draw to date of diagnosis (OR = 2.84, 95% CI = 1.08 to 7.46; P_{trend} = .02 for shorter follow-up and OR = 2.39, 95% CI = 0.76 to 7.55; P_{trend} =.32 for longer follow-up). A stronger association between serum insulin concentration and the risk of prostate cancer was also observed among men who did not receive the ATBC Trial β-carotene supplement than among those who did (for highest vs lowest insulin quartile, OR = 4.21, 95% CI = 1.33 to 13.40, in the no β-carotene supplement arm; and OR = 1.59, 95% CI = 0.55 to 4.61, in the β-carotene arm). However, associations were similar in the vitamin E supplementation arm (OR = 2.43, 95% CI = 0.83 to 7.12) and the no vitamin E supplement arm (OR = 2.57, 95% CI = 0.84 to 7.92) for highest vs lowest insulin quartiles. Similar associations were observed for the HOMA-IR index in subgroup analyses. It should be noted, however, that study power was limited for all exploratory analyses presented in Table 4, and so these results should be viewed with caution.

Discussion

We found an association between higher fasting serum insulin concentrations and an increased risk of developing prostate cancer in the 5–12 years after blood collection. This relation showed a stronger dose–risk trend for lower-stage prostate malignancies. Both the HOMA-IR, a calculated insulin resistance index that is highly dependent on fasting insulin, and the molar ratio of insulin to glucose were associated with the risk of prostate cancer, whereas the fasting glucose concentration was not. These findings support a role for higher circulating insulin in prostate carcinogenesis, especially in early-stage disease.

Clinical studies of prostate cancer have established that insulin concentrations are higher among patients with more advanced disease, greater tumor volume, and higher tumor grade (14,20). To minimize the effects of preexisting clinical or subclinical malignancy and the resulting potential for study bias from reverse causality, we analyzed only case subjects who were diagnosed with prostate cancer at least 5 years (average = 9.2 years) after the baseline blood collection. Three previous cohort investigations did not find an association between circulating insulin concentration and prostate cancer risk; these studies had average times from blood collection to diagnosis of 3.3 years (7), 3.4 years (9), and 13.0 years (8). In addition to the inclusion of case subjects that occurred soon after baseline blood collection and shorter overall study follow-up time, findings from these investigations were based on substantially higher insulin levels and quartile categories than those in this analysis. For example, average serum or plasma insulin concentrations among control subjects in those three studies were 7.9 μU/mL (7), 9.6 μU/mL (case subjects and control subjects combined) (8), and 16.3 μU/mL (9), with insulin concentrations for the quartile categories of 2.9, 5.0, 6.8, or 17.6 μU/mL (means) (7); less than 6.0, 6.0–8.2, 8.3–11.9, or 12 μU/mL or more (ranges) (8); and 5–9, 10–12, 13–17, or 18 μU/mL or more (ranges) (9). In this investigation, the overnight fasting median levels among noncase subjects were 5.1 μU/mL with quartiles of 2.75 or less, more than 2.75 to less than or equal to 4.10, more than 4.10 to less than or equal to 6.10, or more than 6.10 μU/mL. It is therefore possible that elevated insulin levels in the previous cohort populations, whether accounted for by some nonfasting samples or other factors, resulted in examining prostate cancer risk associations in an overall higher range of circulating insulin, without sufficiently low reference categories, which could have contributed to their null findings. The relatively low fasting insulin concentrations that we found to be associated with the lowest risk of developing prostate cancer may provide the smallest proliferative or anabolic stimulus to the prostate and other organs such as the pancreas (16). Our observation that the dose–response

Table 4. Associations between baseline serum insulin level and risk of prostate cancer, adjusted for age and body mass index and stratified by selected baseline characteristics: Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study*

	Insulin quartile					
Subgroup	Quartile 1 1 (≤2.75 μU/mL)	Quartile 2 2 (>2.75 to ≤4.10 μU/mL)	Quartile 3 3 (>4.10 to ≤6.10 μU/mL)	Quartile 4 4 (>6.10 μU/mL)	P_{trend}†	$P_{interaction}$†
Age, y						
<56‡	4/51	8/50	8/55	10/42		
OR (95% CI)	1.00 (ref)	1.80 (0.48 to 6.83)	2.05 (0.52 to 8.02)	3.36 (0.74 to 15.37)	.13	
≥56‡	13/49	15/51	19/48	23/54		.56
OR (95% CI)	1.00 (ref)	1.23 (0.53 to 2.90)	1.79 (0.75 to 4.24)	2.28 (0.91 to 5.70)	.07	
BMI, kg/m²						
<26.1‡	13/78	20/69	13/36	9/15		
OR (95% CI)	1.00 (ref)	2.05 (0.91 to 4.57)	2.46 (0.98 to 6.18)	3.89 (1.31 to 11.54)	.01	
≥26.1‡	4/22	3/32	14/67	24/81		.06
OR (95% CI)	1.00 (ref)	0.53 (0.11 to 2.63)	1.02 (0.30 to 3.50)	1.27 (0.37 to 4.36)	.33	
Occupational activity						
Low‡	10/65	18/64	24/80	32/75		
OR (95% CI)	1.00 (ref)	2.12 (0.89 to 5.04)	2.31 (0.98 to 5.44)	3.66 (1.48 to 9.04)	.04	
High‡	7/35	5/37	3/23	1/21		.04
OR (95% CI)	1.00 (ref)	0.71 (0.20 to 2.53)	0.80 (0.17 to 3.71)	0.35 (0.03 to 4.00)	.42	
No. of cigarettes per day						
<20‡	11/30	5/34	8/35	10/29		
OR (95% CI)	1.00 (ref)	0.47 (0.14 to 1.57)	1.02 (0.33 to 3.17)	1.64 (0.53 to 5.08)	.21	
≥20‡	6/70	18/67	19/68	23/67		.94
OR (95% CI)	1.00 (ref)	3.21 (1.18 to 8.78)	3.16 (1.13 to 8.81)	3.79 (1.23 to 11.74)	.08	
Total energy, kcal/d						
<2721‡	6/51	10/51	12/46	16/44		
OR (95% CI)	1.00 (ref)	1.96 (0.64 to 6.00)	2.61 (0.85 to 8.06)	4.09 (0.24 to 13.45)	.02	
≥2721‡	10/43	12/46	14/53	13/49		.63
OR (95% CI)	1.00 (ref)	1.24 (0.48 to 3.24)	1.29 (0.49 to 3.41)	1.44 (0.47 to 4.41)	.56	
Dietary protein, g/d						
<100‡	8/53	8/54	12/44	12/41		
OR (95% CI)	1.00 (ref)	1.14 (0.39 to 3.36)	2.43 (0.84 to 6.98)	3.21 (0.99 to 10.42)	.04	
≥100‡	8/41	14/43	14/55	17/52		.69
OR (95% CI)	1.00 (ref)	1.82 (0.68 to 4.90)	1.30 (0.47 to 3.62)	1.75 (0.58 to 5.27)	.48	
Dietary carbohydrate, g/d						
<294‡	5/48	10/53	12/51	17/39		
OR (95% CI)	1.00 (ref)	2.41 (0.73 to 7.96)	2.86 (0.85 to 9.59)	6.20 (1.72 to 22.35)	.005	
≥294‡	11/46	12/44	14/48	12/54		.21
OR (95% CI)	1.00 (ref)	1.21 (0.48 to 3.06)	1.37 (0.53 to 3.53)	1.19 (0.40 to 3.57)	.81	
Alcohol intake, g/d						
<11.4‡	11/45	11/48	15/49	12/49		
OR (95% CI)	1.00 (ref)	1.02 (0.39 to 2.63)	1.46 (0.57 to 3.71)	1.15 (0.39 to 3.38)	.89	
≥11.4‡	5/49	11/49	11/50	17/44		.15
OR (95% CI)	1.00 (ref)	2.66 (0.83 to 8.55)	2.42 (0.73 to 8.11)	5.70 (1.60 to 20.28)	.01	
Time to diagnosis, y						
<9.2‡	11/100	9/101	13/103	17/96		
OR (95% CI)	1.00 (ref)	0.96 (0.37 to 2.45)	1.57 (0.63 to 3.88)	2.84 (1.08 to 7.46)	.02	
≥9.2‡	6/100	14/101	14/103	16/96		.45
OR (95% CI)	1.00 (ref)	2.47 (0.90 to 6.78)	2.17 (0.76 to 6.19)	2.39 (0.76 to 7.55)	.32	
Stage						
0–II‡	10/100	14/101	21/103	24/96		
OR (95% CI)	1.00 (ref)	1.62 (0.68 to 3.90)	2.36 (1.00 to 5.56)	3.19 (1.25 to 8.13)	.02	
III–IV‡	7/100	9/101	6/103	8/96		0.38
OR (95% CI)	1.00 (ref)	1.38 (0.49 to 3.91)	0.98 (0.30 to 3.21)	1.53 (0.44 to 5.35)	.57	

* BMI = body mass index (weight [kg]/height [m²]); CI = confidence interval; OR = odds ratio; ref = referent.

† *T* statistic. All statistical tests were two-sided.

‡ No. of case subjects/No. of noncase subjects.

association may have been limited to leaner men (ie, body mass index of <26 kg/m²) is similar to that observed among men with lower waist to hip ratios in two previous studies (8,10) and is consistent with the hypothesis that circulating insulin in the normal, non–insulin resistant range has an effect on prostate cancer risk.

In the most recent prospective study (11) of the insulin surrogate C-peptide, an inverse association between concentrations of C-peptide and prostate cancer risk was reported that was stronger among younger men and for nonaggressive disease that was diagnosed after at least 6.2 years of follow-up. The association was

highly dependent on adjustment for serum leptin, however, and the unadjusted risk estimates for insulin and leptin were nearly identical. Whether fasting serum insulin and nonfasting C-peptide levels are differently associated with prostate cancer risk and how nonfasting C-peptide levels could have interacted with leptin are not clear. These associations should be evaluated further in other prospective investigations.

Our findings are empirically consistent with a population-based, retrospective case–control study (10) in China that reported a relative risk of 2.6 for men with fasting insulin of more than 8.8 μU/mL (compared with those whose fasting insulin level was <6.4 μU/mL). This study primarily examined case subjects with advanced prostate cancer who had a prostate-specific antigen level of higher than 10 ng/mL, however, which may have contributed to the relatively high insulin levels among case subjects (mean = 12.0 vs 7.9 μU/mL in control subjects) (14,20). These concentrations are also somewhat high relative to the low average body mass index (21.7 kg/m^2) and insulin to glucose ratio (ie, 0.09–0.10) of the population compared with the population in this study (ie, means of 26.5 kg/m^2 and 0.04, respectively). Given the retrospective design, both reverse causality and possibly some nonfasting samples are potential explanations for the higher circulating insulin levels reported. It is possible that the similar estimates of the association between serum insulin level and risk of prostate cancer in that case–control study and in our analysis represent two distinct effects that are related to prostate cancer: the first effect being an etiological role for higher chronic circulating insulin, as we demonstrated by the strong relationship between fasting levels and a subsequent diagnosis of prostate cancer 5–12 years later, and the second effect being reverse causality in the retrospective study (10), with later-stage disease among case subjects leading to hyperinsulinemia and consequently a strong positive association between serum insulin level and prostate cancer.

Any etiological relationship between circulating insulin level and prostate cancer risk could also be reflected in studies of the risk associated with diabetes. Type 2 diabetes results in chronic hyperinsulinemia in early stages of the disease followed by hypoinsulinemia in later stages that is induced by pancreatic beta cell exhaustion (21,22). Two recent meta-analyses (23,24) and one recent large cohort study (25) found that being diagnosed with diabetes was associated with a decreased risk of prostate cancer [RR = 0.91, 95% CI = 0.86 to 0.96 (23); RR = 0.84, 95% CI = 0.76 to 0.93 (24); and RR = 0.71, 95% CI = 0.66 to 0.76 (25)]. In theory then, the mechanism of the association between diabetes and a decreased risk of prostate cancer could involve insulin depletion.

Our exploratory subgroup findings offer some areas that should be evaluated in future investigations of the association between serum insulin level and prostate cancer risk. We observed a stronger influence of insulin on early-stage prostate cancer than on advanced disease. When this result is considered with those from the Physicians' Health Study (26) that found reduced prostate cancer survival among men with high prospective C-peptide concentrations, one could hypothesize that hyperinsulinemia may be involved in the development of new prostate cancers as well as in tumor progression and prostate cancer survival. This interpretation is not inconsistent with insulin's stimulation of cell division (27), given the importance of the latter to tumorigenesis, tumor growth, and cancer progression. The stronger insulin association among men with body mass index of less than 26 kg/m^2 (median for the category = 23 kg/m^2) may also represent a more isolated effect of fasting insulin on prostate epithelial mitogenesis, growth, and subsequent carcinogenesis that is not influenced or confounded by the lower circulating testosterone, sex hormone–binding globulin, growth hormone, and insulin-like growth factor I or higher estrogens that result from obesity and increased visceral fat (28). As mentioned above, this interaction has been observed in two previous studies (8,10) and could also be due to insulin's reduction of sex hormone–binding globulin synthesis (29) (with resulting higher levels of free testosterone) in the setting of elevated sex hormone–binding globulin levels among men with lower body mass index. The fact that adjustment for body mass index did not alter the risk estimates for the association between serum insulin concentration and prostate cancer risk in our study also supports the possibility that its influence is independent of being overweight or obese and indicates that its direct effects on the prostate epithelium may be more relevant than insulin resistance. The interactions with lower occupational activity, heavier smoking, lower energy intake, and greater alcohol consumption may or may not represent real biological modulation of circulating insulin or may be because of chance; however, they should be evaluated in other investigations.

Insulin is essential for normal human growth, development, and uptake of glucose and amino acids as well as increased protein and fatty acid synthesis by cells throughout the body including the prostate. Insulin is also a potent growth factor that acts by binding to cell-membrane insulin receptors and stimulating mitosis through protein kinase B/Akt-mediated signal transduction and DNA synthesis; insulin also has antiapoptotic properties (27). In addition, greater Akt activation, increased insulin receptor levels, and increased LNCaP tumor growth and aggressiveness have been reported in response to dietary modification, which increased plasma insulin in mice (27). Greater expression of insulin receptor isoforms in human prostate cancers and observations of increased insulin receptor levels, as shown by immunohistochemical staining, in higher grade lesions also supports the possibility that prostate cancer tissue can respond to changes in insulin levels (30). Such studies provide experimental evidence for the biological impact of higher circulating insulin both on incident prostate cancers (eg, through anabolic and mitogenic stimulation) and on prostate cancer progression (eg, through increased insulin–insulin receptor binding in prostate cancer cell membranes and resulting tumor cell proliferation).

Our investigation of fasting serum insulin is limited insofar as the sample size was not large, and it included only Finnish male smokers (all whites) who were originally enrolled in a cancer prevention trial of vitamin supplementation. The findings may or may not, therefore, be generalizable to other populations. Examination of the hypothesis that fasting serum insulin is etiologically related to prostate cancer in nonsmoking populations and other races/ethnicities would be particularly useful, for example. Several aspects of study design, however, including the population-based cancer registry ascertainment of case subjects, availability of prospectively collected overnight fasting serum and of other measured exposures (eg, body mass index), relatively long follow-up, and

laboratory quality control procedures, should have afforded a valid estimation of the association between serum insulin level and prostate cancer risk.

Although the relationship of obesity and the insulin resistance syndrome with prostate cancer risk is somewhat complex because of the associated biological changes (eg, circulating androgens and insulin-like growth factor I) and potential effects on detection of early-stage disease (31,32), results from this study support an association between circulating insulin levels and prostate cancer risk that appears to be independent of relative weight and possibly insulin resistance. Our investigation also highlights the importance and relevance of actual absolute biomarker concentrations (in this case, serum insulin) and population distributions, as well as fasting vs nonfasting blood collection, when comparing the results across multiple studies. Future similar prospective investigations of fasting insulin that exclude several years of prostate cancer cases diagnosed early following blood collection would be informative and help provide a more complete picture regarding the insulin–prostate cancer "dose–risk" relation and stages of disease that are impacted.

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