

Insulin, Glucose, Insulin Resistance, and Incident Colorectal Cancer in Male Smokers

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Background & Aims: Hyperinsulinemia is a putative colorectal cancer (CRC) risk factor. Insulin resistance (IR) commonly precedes hyperinsulinemia and can be quantitatively measured by using the homeostasis model assessment–insulin resistance (HOMA-IR) index. To date, few studies have directly examined serum insulin as an indicator of CRC risk, and none have reported associations on the basis of HOMA-IR.

Methods: We performed a case-cohort study within the Alpha-Tocopherol, Beta-Carotene Cancer Prevention (ATBC) Study (n = 29,133). Baseline exposure and fasting serum biomarker data were available for 134 incident CRC case and 399 non-case subjects. HOMA-IR was derived as fasting insulin × fasting glucose/22.5. Hazard ratios (HRs) and 95% confidence intervals (CIs) were estimated by using age-adjusted and multivariable-adjusted Cox proportional hazards regression models. **Results:** Median (interquartile range) values for serum insulin, glucose, and HOMA-IR were 4.1 (2.9–7.2) mIU/L, 101 (94–108) mg/dL, and 0.99 (0.69–1.98) for case subjects and 4.1 (2.7–6.1) mIU/L, 99 (93–107) mg/dL, and 1.02 (0.69–1.53) for non-case subjects, respectively. On the basis of comparison of the highest versus lowest quartiles for each biomarker, insulin (HR, 1.84; 95% CI, 1.03–3.30) and HOMA-IR (HR, 1.85; 95% CI, 1.06–3.24) were significantly associated with incident CRC, whereas glucose was marginally associated with incident CRC (HR, 1.70; 95% CI, 0.92–3.13) in age-adjusted risk models. However, trends across biomarker quartiles were somewhat inconsistent (*P* trend = .12, .04, and .12, respectively), and multivariable adjustment generally attenuated the observed risk estimates. **Conclusions:** Data from this prospective study of male smokers provide limited support for hyperinsulinemia, hyperglycemia, and/or insulin resistance as CRC risk factors.

Data from a variety of sources suggest that insulin might play a functional role in colorectal carcinogenesis.^{1–3} Insulin administration stimulates proliferation and reduces apoptosis in colorectal cancer (CRC) cell lines^{4–6} and also promotes colorectal tumor growth in animal model systems.^{7–9} In addition, multiple epidemiologic studies have reported positive associations between type 2 diabetes mellitus (DM) and CRC risk, as recently reviewed.^{3,10} Because type 2 DM is characterized by increased circulating insulin concentration during the early stages of disease,¹¹ these reports indirectly support the “hyperinsulinemia hypothesis.” However, to date, few studies have directly examined serum or plasma insulin level as a CRC risk factor.^{12–15}

Insulin resistance (IR), defined as a subnormal glycemic response to endogenous insulin, precedes hyperinsulinemia

among type 2 DM patients¹¹ and has been proposed as the primary mediator of increased CRC risk among obese individuals.^{1,16} IR is most accurately measured by using the hyperinsulinemic-euglycemic clamp technique,¹⁷ but this method is impractical for large-scale epidemiologic studies. Several IR indices can be derived from fasting serum insulin and glucose levels, such as the homeostasis model assessment (HOMA-IR).^{18,19} HOMA-IR has been positively associated with cancer risk outside of the colorectum,^{20,21} but no data have been reported with respect to HOMA-IR as an indicator of CRC risk.

In this prospective case-cohort study, we evaluated associations between baseline serum insulin, glucose, and HOMA-IR levels with incident CRC among a subset of Finnish male smokers enrolled in the Alpha-Tocopherol, Beta-Carotene Cancer Prevention (ATBC) Study. Because cigarette smoking has been shown to induce IR²² and is also a putative CRC risk factor,²³ we anticipated that investigation of the proposed risk associations might be particularly informative in this subject population. Also, because proximal and distal CRCs exhibit distinct molecular, histologic, and clinical features,^{24–26} we estimated CRC risks overall and by anatomic subsite for each of the measured serum biomarkers.

Materials and Methods

Details regarding design and conduct of the ATBC Study have been previously described.²⁷ In brief, 29,133 men ages 50–69 years who lived in southwestern Finland and smoked at least 5 cigarettes per day were recruited between 1985–1988. Individuals with a previous cancer history (except non-melanoma skin cancer) were excluded. Enrolled trial participants provided a fasting blood sample before randomization, from which serum specimens were isolated, aliquoted, and stored deep-frozen at –70°C for future analyses. Intervention groups were randomly assigned on the basis of a complete 2 × 2 factorial study design (alpha-tocopherol alone, beta-carotene alone, both, or placebo). Written informed consent was obtained from all trial participants before randomization, and the

Abbreviations used in this paper: ATBC, Alpha-Tocopherol, Beta-Carotene Cancer Prevention; BMI, body mass index; CI, confidence interval; CRC, colorectal cancer; DM, diabetes mellitus; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment–insulin resistance; HR, hazard ratio; ICD-9, International Classification of Diseases-ninth revision; IGF, insulin-like growth factor; IR, insulin resistance; OR, odds ratio; RR, relative risk.

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study was approved by the institutional review boards of both the National Public Health Institute in Finland and the U.S. National Cancer Institute.

Incident cancers in the ATBC Study cohort have been identified through the Finnish Cancer Registry, which provides nearly 100% case ascertainment.²⁸ To facilitate efficient serum biomarker studies, a subcohort ($n = 400$) of randomly selected trial participants was assembled from among all subjects who were alive and without a cancer diagnosis during the first 5 years of cohort follow-up. Subjects who developed incident cancer in one of several target organs, including the colorectum, after their fifth year of cohort follow-up through December 31, 1997 were then selected for batch analyses of serum glucose and insulin levels. In the present study, we included CRC case subjects for whom complete insulin and glucose data were available ($n = 134$). One subject in the subcohort was diagnosed with incident CRC during the first 5 years of cohort follow-up and was dropped from the non-case subcohort. Therefore, our final analytic cohort included 134 CRC case subjects and 399 non-case subjects. CRC diagnoses were confirmed by independent review of all relevant medical records by 2 study physicians (International Classification of Diseases-ninth revision [ICD-9] codes 153.0-153.4, 153.6-153.9, and 154.0-154.1). The interval between serum collection and follow-up was up to 12 years (median follow-up time for incident CRC diagnosis was 9 years).

Demographic, anthropometric, and exposure data were obtained from the ATBC Study baseline questionnaire and physical examination. Variables of interest for the present analyses included age at randomization, height, weight, body mass index (BMI; kg/m^2), systolic and diastolic blood pressure, occupational and recreational physical activity levels, history of DM, cigarette pack-years, education level, urban residence, dietary intake (total energy, carbohydrates, protein, fat, fiber, folate, and calcium), alcohol consumption, and trial intervention group. Baseline serum total and high-density lipoprotein (HDL) cholesterol levels had been previously measured²⁷ and were included in the current analyses as well.

Serum glucose and insulin concentrations were analyzed at Mayo Clinic Rochester by experienced laboratory personnel without prior knowledge of case status. Glucose was measured on the Hitachi 912 Chemistry Analyzer (Hitachi High-Tech Science Systems Corp, Ibaraki, Japan) by using the hexokinase reagent from Boehringer Mannheim (Indianapolis, IN). Insulin level was determined by using a specific 2-site immunoenzymatic assay performed on the Access automated immunoassay system (Beckman Instruments, Chaska, MN) that has a molar cross-reactivity of 0.10% with proinsulin. Serum sets included case, control, and quality control samples. On the basis of results obtained with the quality control samples, within-batch coefficients of variation were 1.1% for the serum glucose assay and 3.5% for the serum insulin assay. Between-batch coefficients of variation for the serum glucose and serum insulin assays were 2.2% and 3.6%, respectively. HOMA-IR was derived as follows: $\text{fasting insulin} \times \text{fasting glucose}/22.5$.

Distributions of demographic and clinical attributes were compared by case status by using χ^2 tests for categorical variables and Wilcoxon rank sum tests for continuous variables. For these analyses, data were descriptively displayed by using frequencies and percents for categorical variables and medians and interquartile ranges for continuous variables. The same set

of covariates were compared across quartiles of the serum biomarkers by using analyses of covariance for continuous variables, ordinary logistic regression analyses for binary variables, and multicategorical nominal logistic regression analyses for all other categorical variables. Analyses were subset to participants in the original subcohort and were adjusted for age. Data were descriptively displayed by using age-adjusted means and percents, as appropriate, with corresponding 95% confidence limits. *P* values were calculated with tests for trend, assuming an inherent ordering of quartiles from lowest to highest. Pairwise associations of serum biomarkers were assessed with Spearman correlation coefficients. Cox proportional hazards regression models were fit to evaluate associations between the serum biomarkers and case status by using methods outlined by Prentice²⁹ to account for the case-cohort study design. We used a robust variance estimate based on the infinitesimal jackknife approach to account for the oversampling of cases.^{30,31} For all Cox analyses, we modeled survival as a function of age, because age is a better predictor of CRC risk in this study than follow-up time.³²

Separate analyses were carried out for insulin, glucose, and HOMA-IR. The serum biomarker data were categorized into approximate quartiles on the basis of the distribution of each variable within the nested subcohort, with the lowest quartile assigned as the reference group. *P* values were again calculated with a one degree-of-freedom test for trend. Age-adjusted and multivariable-adjusted risk associations were assessed for incident CRC overall, as well as for proximal (ICD-9 codes 153.0, 153.1, 153.4, 153.6, 153.7) and distal (ICD-9 codes 153.2, 153.3, 154.0, 154.1) tumors. Multivariable models were developed by adding potential confounders individually into the base model. Age at randomization and cigarette pack-years were included in all multivariable models, because all subjects were smokers, and smoking is a putative CRC risk factor.²³ Other variables listed in Table 1 were included in the final model if any of the following criteria were met for serum insulin, glucose, or HOMA-IR: univariately associated with both the exposure and the outcome ($P < .05$); inclusion changed the serum biomarker hazard ratio by at least 10%; associated with a *P* value of $\leq .20$ in the age- and smoking-adjusted risk model; or inclusion decreased the standard error for any of the serum biomarker risk estimates.

Effect modification of the insulin, glucose, and HOMA-IR risk associations by total energy intake and factors associated with IR syndrome (BMI, occupational physical activity, recreational physical activity, hypertension, total cholesterol, and HDL cholesterol) were investigated by using cross-product terms in multivariable-adjusted Cox regression models. The serum biomarker trend variables were included in all such models to assess differences in the dose-response relationship across levels of the potential effect modifiers. All statistical tests were performed 2-sided, with analyses carried out by using the SAS (SAS Institute, Inc, Cary, NC) and S-Plus (Insightful, Inc, Seattle, WA) software systems.

Results

Selected baseline characteristics of the CRC case and subcohort non-case subjects are shown in Table 1. CRC case subjects were slightly older ($P < .001$) and less physically active at work ($P = .006$) than the non-case subjects. Median (interquartile range) values for serum insulin, glucose, and HOMA-IR were 4.1 (2.9-7.2) mIU/L, 101 (94-108) mg/dL, and 0.99 (0.69-1.98) for case subjects and 4.1 (2.7-6.1) mIU/L, 99 (93-107)

Table 1. Baseline Characteristics of Case-Cohort Study Participants, by Case Status

Characteristic ^a	Incident CRC cases (N = 134)	Subcohort non-cases (N = 399)	P value ^b
Demographics			
Age at randomization (y)	59 (55–62)	56 (52–60)	<.001
BMI (kg/m ²)	26.6 (24.0–29.8)	26.2 (23.9–28.9)	.39
Primary school education or less (N) (%)	108 (81)	313 (78)	.59
Urban residence (N) (%)	89 (66)	269 (67)	.83
Cigarette pack-years	32 (22–44)	35 (25–45)	.27
History of DM (N) (%)	3 (2)	18 (5)	.24
Hypertension at baseline (N) (%) ^c	96 (72)	257 (64)	.13
Dietary intake			
Total energy (kcal/d)	2704 (2222–3253)	2721 (2314–3256)	.62
Carbohydrates (g/d)	293 (229–346)	294 (240–360)	.43
Protein (g/d)	98 (81–118)	100 (83–121)	.70
Fat (g/d)	115 (93–150)	115 (95–148)	.80
Fiber (g/d)	23.1 (18.5–29.7)	24.3 (18.4–31.5)	.44
Folate (mg/d) ^d	327 (250–396)	327 (268–399)	.52
Calcium (mg/d) ^d	1311 (969–1696)	1370 (1028–1729)	.28
Alcohol (g/d)	10.7 (2.8–27.4)	11.4 (2.2–26.9)	.55
Physical activity			
Occupational (N) (%)			.006
Sedentary	22 (16)	54 (14)	
Light/moderate	35 (26)	161 (40)	
Heavy	8 (6)	36 (9)	
Non-working	69 (52)	148 (37)	
Recreational (N) (%)			.70
Sedentary	52 (39)	171 (43)	
Light/moderate	74 (55)	204 (51)	
Heavy	8 (6)	24 (6)	
Serum cholesterol			
Total (mmol/L)	6.1 (5.4–6.8)	6.3 (5.5–7.1)	.17
HDL (mmol/L)	1.11 (0.95–1.37)	1.16 (0.98–1.37)	.28

^aMedian (interquartile range) unless otherwise indicated.

^bBased on χ^2 test (categorical variables) or Wilcoxon rank sum test (continuous variables).

^cDefined as systolic blood pressure >140 mm Hg or diastolic blood pressure >90 mm Hg.

^dIncluding supplements.

mg/dL, and 1.02 (0.69–1.53) for non-case subjects, respectively. Age-adjusted baseline characteristics of the nested subcohort are provided by quartile of HOMA-IR in Table 2. BMI, history of DM, hypertension at baseline, protein intake, fat intake, and calcium intake were positively associated with HOMA-IR, whereas alcohol intake, recreational physical activity, and HDL cholesterol were inversely associated with this insulin resistance biomarker ($P < .05$ for each variable). Serum insulin and glucose levels also increased progressively across HOMA-IR quartiles (P trend < .001 for each biomarker), and correlations between the serum biomarkers were strong: insulin:glucose ($r = 0.44$), insulin:HOMA-IR ($r = 0.98$), and glucose:HOMA-IR ($r = 0.58$).

In age-adjusted risk models (Tables 3 through 5), serum biomarker levels in the highest versus lowest quartiles were associated with increased CRC risk for insulin (hazard ratio [HR], 1.84; 95% confidence interval [CI], 1.03–3.30), glucose (HR, 1.70; 95% CI, 0.92–3.13), and HOMA-IR (HR, 1.85; 95% CI, 1.06–3.24). The trends across quartiles were somewhat inconsistent, however (P trend = .12, .04, and .12, respectively). Multivariable adjustment generally attenuated the observed risk associations, with slightly lower risk estimates for each extreme quartile comparison and absence of statistically significant trends across quartiles: insulin (HR, 1.74; 95% CI, 0.74–4.07; P trend = .40), glucose (HR, 1.65; 95% CI, 0.78–3.49; P trend =

.16), and HOMA-IR (HR, 1.71; 95% CI, 0.77–3.78; P trend = .38). Further analyses based on proximal and distal CRC subsites did not reveal any material differences in the associations with insulin, glucose, or HOMA-IR (Tables 3 through 5). Consideration of rectal cancers separately from distal colon cancers also did not appreciably alter the subsite-specific risk estimates (data not shown). No statistically significant effect modification on the serum biomarker risk associations was detected from total energy intake, BMI, hypertension, occupational physical activity, recreational physical activity, total cholesterol, or HDL cholesterol level ($P > .05$ for each comparison).

Discussion

In this prospective study of Finnish male smokers, baseline fasting insulin and HOMA-IR were positively associated with incident CRC in age-adjusted risk models. Glucose was also associated with increased CRC risk, but the age-adjusted risk estimate did not achieve statistical significance. These data add to the limited number of published reports wherein circulating insulin and/or glucose concentrations have been directly examined in relation to incident CRC. We also provide novel data regarding HOMA-IR as an indicator of CRC risk. These data support the hypothesis that hyperinsulinemia, hyperglycemia, and/or insulin resistance might be functionally

Table 2. Age-Adjusted Baseline Characteristics of Subcohort Non-Case Subjects, by Quartile of HOMA-IR

Characteristic	HOMA-IR				P trend
	Quartile 1	Quartile 2	Quartile 3	Quartile 4	
Demographics					
Age at randomization (y) ^a	56.5 (55.5–57.4)	56.3 (55.3–57.2)	56.0 (55.0–57.0)	56.7 (55.7–57.7)	.81
BMI (kg/m ²) ^b	24.3 (23.6–24.9)	25.2 (24.5–25.8)	27.1 (26.5–27.8)	29.9 (29.2–30.5)	<.001
Primary school education or less ^c	82.4 (75.0–89.8)	79.9 (72.0–87.8)	68.8 (59.6–78.0)	84.7 (77.7–91.7)	.78
Urban residence ^c	73.0 (61.4–79.1)	63.3 (53.8–72.8)	72.2 (63.4–81.1)	62.2 (52.8–71.7)	.47
Cigarette pack-years ^b	36 (33–39)	38 (34–41)	33 (30–36)	37 (34–41)	.95
Self-reported DM ^c	0	0.8 (0–2.6)	2.4 (0–5.4)	15.0 (8.0–21.9)	<.001
Hypertension at baseline ^c	50.3 (40.6–60.0)	65.6 (56.2–74.9)	66.7 (57.4–76.0)	76.6 (68.4–84.9)	<.001
Dietary intake (per day)^{b,d}					
Total energy (kcal)	2853 (2689–3018)	2779 (2614–2944)	2816 (2648–2983)	2893 (2729–3057)	.68
Carbohydrates (g)	308 (299–317)	310 (301–319)	303 (294–312)	298 (289–307)	.08
Protein (g)	101 (98–103)	102 (100–105)	105 (102–108)	107 (104–110)	.001
Fat (g)	121 (117–125)	120 (117–124)	125 (121–128)	127 (124–131)	.007
Fiber (g)	26.2 (24.6–27.7)	25.8 (24.2–27.3)	25.3 (23.8–26.9)	25.0 (23.4–26.9)	.24
Folate (mg)	328 (316–340)	345 (334–357)	345 (333–357)	346 (334–357)	.05
Calcium (mg)	1380 (1289–1471)	1377 (1286–1469)	1443 (1351–1536)	1585 (1495–1677)	.001
Alcohol (g)	23.0 (18.4–27.7)	20.8 (16.2–25.5)	17.8 (13.1–22.5)	16.8 (12.2–21.4)	.04
Physical activity					
Occupational^c					
Sedentary	10.8 (4.8–16.9)	12.6 (6.0–19.1)	14.5 (7.6–21.5)	14.0 (7.2–20.8)	.16
Light/moderate	39.7 (30.2–49.2)	39.7 (30.1–49.4)	42.0 (32.2–51.7)	33.4 (24.2–42.6)	
Heavy	11.5 (5.3–17.7)	11.2 (5.0–17.4)	6.6 (1.7–11.6)	5.4 (1.0–9.8)	
Non-working	38.0 (28.5–47.4)	36.5 (27.0–46.0)	36.9 (27.3–46.4)	47.2 (37.5–56.9)	
Recreational^c					
Sedentary	42.4 (32.8–52.0)	37.4 (27.9–47.0)	35.6 (26.1–45.0)	54.7 (45.0–64.4)	.04
Light/moderate	46.2 (36.5–55.9)	57.4 (47.6–67.1)	59.1 (49.4–68.9)	42.2 (32.6–51.9)	
Heavy	11.9 (5.2–17.6)	5.2 (0.8–9.5)	5.3 (0.9–9.8)	3.1 (0–6.5)	
Serum biomarkers					
Total cholesterol (mmol/L) ^b	6.2 (6.0–6.4)	6.3 (6.1–6.5)	6.6 (6.4–6.8)	6.3 (6.1–6.5)	.21
HDL cholesterol (mmol/L) ^b	1.34 (1.28–1.40)	1.26 (1.20–1.32)	1.18 (1.12–1.24)	1.06 (1.00–1.12)	<.001
Insulin (mIU/mL) ^b	2.08 (1.54–2.63)	3.53 (2.97–4.08)	5.00 (4.44–5.55)	9.97 (9.42–10.52)	<.001
Glucose (mg/dL) ^b	92 (88–96)	99 (95–103)	102 (98–107)	121 (116–125)	<.001

^aUnadjusted analysis of variance.^bAnalyses of covariance, adjusting for age; mean (95% CI).^cBinary and multicategorical logistic regression analyses, adjusting for age; percent (95% CI).^dIn addition to age, dietary variables were also adjusted for energy, with the exception of alcohol; folate and calcium intake include supplements.

involved in colorectal carcinogenesis. However, given the somewhat inconsistent CRC risk estimates observed across biomarker quartiles, we speculate that serial analyses of fasting and non-fasting serum samples might permit more accurate characterization of chronic insulin and glucose exposures.

Previous studies of circulating insulin concentration (either fasting or non-fasting) and CRC risk have yielded inconsistent results. In the Cardiovascular Health Study, fasting insulin levels did not show a linear relationship with incident CRC (statistical test not provided)¹²; however, insulin levels above the cohort median were associated with increased CRC risk in secondary analyses (relative risk [RR], 1.6; 95% CI, 1.1–2.4). Furthermore, insulin levels obtained 2 hours after a 75-g oral glucose load were associated with a 2-fold CRC risk elevation (RR, 2.0; 95% CI, 1.0–3.8 for comparison of extreme quartiles; *P* trend = .04). Conversely, in a nested case-control study from northern Sweden,¹³ non-fasting insulin levels were actually higher among controls than cases, with mean values of 81.6 and 64.2 pmol/L, respectively (*P* < .05). Adjusting for smoking status in logistic regression models reversed the directionality of the observed association, but the risk estimate remained

statistically nonsignificant (odds ratio [OR], 1.22; 95% CI, 0.64–2.31 for comparison of highest to lowest quartiles; *P* trend = .41). Similarly, a nested case-control study of Washington County, Maryland residents found no significant association between non-fasting insulin levels and incident CRC (OR, 0.78; 95% CI, 0.45–1.35 for comparison of highest to lowest quartiles; *P* trend = .24).¹⁴ More recently, 2 case-control studies reported positive associations between fasting insulin levels and prevalent colorectal adenomas.^{15,33}

C-peptide, which is cleaved from proinsulin, has a relatively long half-life in the peripheral circulation³⁴ and might provide a more accurate assessment of overall insulin exposure (ie, basal plus stimulated levels). In the New York University Women's Health Study,³⁵ CRC risk was increased by approximately 3-fold among subjects with C-peptide levels in the highest versus lowest quintiles (OR, 2.92; 95% CI, 1.26–6.75; *P* trend = .001). Further adjustment for BMI modestly strengthened the observed risk association (OR, 3.28; 95% CI, 1.30–8.26). A nested case-control study of Physicians' Health Study participants³⁶ also found that C-peptide levels were positively associated with incident CRC (RR, 2.7; 95% CI, 1.2–6.2; *P* trend = .05). Sub-

Table 3. Associations Between Fasting Insulin and Incident CRC, Overall and by Anatomic Subsite

	Insulin (IU/mL)				P trend ^a
	Quartile 1 (<2.8)	Quartile 2 (2.9–4.1)	Quartile 3 (4.2–6.1)	Quartile 4 (>6.1)	
All CRC cases/subcohort non-cases (N)	31/104	37/97	25/103	41/95	
Age-adjusted HR (95% CI)	1.00 (ref)	1.40 (0.78–2.51)	0.94 (0.51–1.74)	1.84 (1.03–3.30)	.12
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.43 (0.74–2.78)	0.77 (0.35–1.69)	1.74 (0.74–4.07)	.40
Proximal CRC cases/subcohort non-cases (N)	13/104	13/97	9/103	12/95	
Age-adjusted HR (95% CI)	1.00 (ref)	1.30 (0.56–3.04)	0.91 (0.36–2.33)	1.53 (0.62–3.76)	.54
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.36 (0.53–3.52)	0.82 (0.26–2.65)	1.62 (0.45–5.86)	.64
Distal CRC cases/subcohort non-cases (N)	18/104	24/97	16/103	29/95	
Age-adjusted HR (95% CI)	1.00 (ref)	1.46 (0.72–2.97)	0.96 (0.46–1.99)	2.03 (1.04–3.97)	.10
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.45 (0.65–3.27)	0.73 (0.28–1.87)	1.73 (0.63–4.77)	.49

NOTE. All risk estimates are based on Cox proportional hazards regression analyses, modeling risk as a function of age and accounting for the case-cohort study design.

^aBased on test for trend.

^bAdjusted for cigarette pack-years, BMI, protein intake, fat intake, fiber intake, alcohol consumption, caloric intake, history of DM, and occupational physical activity.

group analyses restricted to subjects whose blood samples were collected after fasting for at least 4 hours revealed a slightly lower, statistically nonsignificant risk estimate. In contrast, C-peptide levels were not associated with CRC risk (RR, 1.17; 95% CI, 0.63–2.20) in a nested case-control study of Nurses' Health Study participants.³⁷

Hyperglycemia has also been evaluated as a potential CRC risk factor, but existing data remain inconclusive. In the Cardiovascular Health Study,¹² subjects in the highest quartile for fasting glucose at baseline were nearly 2 times more likely to develop incident CRC, compared with subjects in the lowest quartile (RR, 1.8; 95% CI, 1.0–3.1; *P* trend = .02). CRC risk was similarly elevated among women (RR, 1.98; 95% CI, 1.31–2.98) but not men (RR, 0.90; 95% CI, 0.58–1.40) with elevated blood glucose levels in a prospective, community-based study from Norway.³⁸ However, 2 other cohort studies from Korea³⁹ and Japan⁴⁰ found no significant associations between fasting serum glucose level and incident CRC. Glycosylated hemoglobin, which provides an indication of average blood glucose levels during the preceding 3 months, was positively associated with CRC risk in the European Prospective Investigation into Can-

cer-Norfolk Study (RR, 1.34; 95% CI, 1.12–1.59 for every 1% absolute increase in hemoglobin A_{1c} level⁴¹ and in the Washington County nested case-control study (OR, 1.57; 95% CI, 0.94–2.60 for comparison of extreme quartiles; *P* trend = .02)¹⁴ but not in the Nurses' Health Study.³⁷ Blood glucose levels after oral glucose challenge have also shown mixed results with respect to predicting CRC risk. Among subjects in the Cardiovascular Health Study,¹² post-challenge glucose levels were associated with a higher CRC risk (RR, 2.4; 95% CI, 1.2–4.7; *P* trend = .02) than were fasting glucose levels (RR, 1.8; 95% CI, 1.0–3.1; *P* trend = .02). Extended follow-up from another prospective study of cardiovascular disease screening program participants⁴² demonstrated a 64% increase in CRC mortality among those with post-challenge glucose levels in the highest versus lowest quartiles (RR, 1.64; 95% CI, 1.13–2.37; *P* trend = .05). In the Second National Health and Nutrition Examination Survey (NHANES II) Mortality Study, impaired glucose tolerance was associated with a strikingly elevated risk for colon cancer mortality (RR, 4.24; 95% CI, 1.25–14.41), although the point estimate was based on relatively few fatal events (*n* = 15).⁴³ Furthermore, a relatively small study from Japan reported

Table 4. Associations Between Fasting Glucose and Incident CRC, Overall and by Anatomic Subsite

	Glucose (mg/dL)				P trend ^a
	Quartile 1 (<92)	Quartile 2 (93–98)	Quartile 3 (99–107)	Quartile 4 (>107)	
All CRC cases/subcohort non-cases (N)	23/99	32/100	44/101	35/99	
Age-adjusted HR (95% CI)	1.00 (ref)	1.35 (0.72–2.50)	2.08 (1.14–3.79)	1.70 (0.92–3.13)	.04
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.19 (0.58–2.43)	1.95 (0.97–3.91)	1.65 (0.78–3.49)	.16
Proximal CRC cases/subcohort non-cases (N)	8/99	14/100	15/101	10/99	
Age-adjusted HR (95% CI)	1.00 (ref)	1.57 (0.62–3.96)	2.15 (0.85–5.41)	1.45 (0.55–3.84)	.33
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.67 (0.59–4.73)	1.92 (0.66–5.58)	1.38 (0.42–4.52)	.52
Distal CRC cases/subcohort non-cases (N)	15/99	18/100	29/101	25/99	
Age-adjusted HR (95% CI)	1.00 (ref)	1.22 (0.58–2.57)	2.04 (1.02–4.09)	1.81 (0.90–3.66)	.04
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.03 (0.42–2.51)	2.09 (0.89–4.91)	1.85 (0.77–4.45)	.08

NOTE. All risk estimates are based on Cox proportional hazards regression analyses, modeling risk as a function of age and accounting for the case-cohort study design.

^aBased on test for trend.

^bAdjusted for cigarette pack-years, BMI, protein intake, fat intake, fiber intake, alcohol consumption, caloric intake, history of DM, and occupational physical activity.

Table 5. Associations Between HOMA-IR and Incident CRC, Overall and by Anatomic Subsite

	HOMA-IR				P trend ^a
	Quartile 1 (<92)	Quartile 2 (93-98)	Quartile 3 (99-107)	Quartile 4 (>107)	
All CRC cases/subcohort non-cases (N)	33/102	37/99	18/98	46/100	
Age-adjusted HR (95% CI)	1.00 (ref)	1.27 (0.71-2.26)	0.67 (0.35-1.30)	1.85 (1.06-3.24)	.12
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.30 (0.68-2.47)	0.51 (0.23-1.16)	1.71 (0.77-3.78)	.38
Proximal CRC cases/subcohort non-cases (N)	14/102	12/99	6/98	15/100	
Age-adjusted HR (95% CI)	1.00 (ref)	1.11 (0.47-2.61)	0.63 (0.22-1.79)	1.70 (0.73-3.97)	.40
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.33 (0.52-3.42)	0.47 (0.13-1.67)	1.96 (0.56-6.91)	.54
Distal CRC cases/subcohort non-cases (N)	19/102	25/99	12/98	31/100	
Age-adjusted HR (95% CI)	1.00 (ref)	1.36 (0.69-2.70)	0.70 (0.32-1.52)	1.94 (1.01-3.73)	.15
Multivariable-adjusted HR (95% CI) ^b	1.00 (ref)	1.31 (0.60-2.87)	0.53 (0.20-1.38)	1.58 (0.63-3.93)	.53

NOTE. All risk estimates are based on Cox proportional hazards regression analyses, modeling risk as a function of age and accounting for the case-cohort study design.

^aBased on test for trend.

^bAdjusted for cigarette pack-years, BMI, protein intake, fat intake, fiber intake, alcohol consumption, caloric intake, history of DM, and occupational physical activity.

that post-challenge glucose levels (OR, 1.41; 95% CI, 1.05-1.88) were positively associated with prevalent colorectal adenomas.⁴⁴ In contrast, Smith et al⁴⁵ found null associations for both colon (RR, 1.03; 95% CI, 0.88-1.24) and rectal (RR, 0.94; 95% CI, 0.70-1.27) cancer mortality among subjects in the highest versus lowest quartiles for post-load glucose levels in a large study of male civil servants from the United Kingdom.

Insulin resistance has been observed to be a risk factor for several chronic conditions, including atherosclerosis, hypertension, dyslipidemia, and non-alcoholic fatty liver disease, as well as extracolonic cancers.^{20,21,46-52} Because HOMA-IR is derived from paired serum insulin and glucose values, this composite index might provide an earlier indication of evolving hyperinsulinemia and/or hyperglycemia. However, CRC risks associated with baseline insulin, glucose, and HOMA-IR were not appreciably different in our study. Because consensus has not been achieved regarding the most appropriate IR index to use for epidemiologic research,^{17,53} we also analyzed CRC risks on the basis of the Quantitative Insulin Sensitivity Check Index (QUICKI). The observed risk estimates were similar to HOMA-IR (data not shown).

The range of insulin values observed in our study was relatively narrow, possibly representing the degree of the overnight fast, but which limited the ability to detect small effect sizes. Because insulin assay techniques are not standardized, comparison of absolute insulin values across studies is largely uninformative.⁵⁴ In the only other prospective study of fasting insulin levels and incident CRC reported to date,¹² distribution of the predictor variable appears to have been more pronounced (range, 4-400 IU/mL in men and 3-400 in women) than observed here, but the association with CRC risk also failed to achieve statistical significance. It is also possible that insulin-like growth factor (IGF) proteins, such as IGF-1 and IGF-binding protein 3, might be more relevant to colorectal carcinogenesis than insulin, glucose, or HOMA-IR. However, existing data remain inconsistent,⁵⁵ and preliminary analyses of IGF-1 and IGF-binding protein 3 levels in our case-cohort study did not reveal any statistically significant associations with incident CRC.⁵⁶

The relatively restricted demographic characteristics of our subject population (ie, all older male smokers) should be taken

into account when interpreting the external validity of these observations. Nonetheless, several strengths of our study are worthy of consideration. First, analyses of serum samples obtained >5 years before incident CRC diagnosis effectively removed the possibility that the serum biomarker levels were influenced by physiologic factors or lifestyle changes induced by subclinical colorectal neoplasia. Second, the identification of CRC cases and controls from within the same source population minimized the chance of selection bias. Third, adjustment for multiple conditions associated with IR syndrome,^{46,57} as well as other potential confounding variables, allowed us to define independent associations between serum insulin, glucose, and HOMA-IR levels with incident CRC. In fact, we might have overadjusted for one or more factors within the causal pathway, because multivariable adjustment generally attenuated the observed risk estimates. As noted above, measurement of insulin and glucose levels from a single, fasting serum sample might not adequately characterize long-term exposure. Several prior observational studies have reported stronger CRC risk associations based on non-fasting or time-averaged indicators of hyperinsulinemia or hyperglycemia, suggesting that stimulated insulin and glucose levels might also be more relevant to colorectal carcinogenesis.

In summary, data from this prospective study support the possibility that aberrant insulin and/or glucose homeostasis, perhaps as a consequence of IR, might be functionally related to CRC risk. In light of the emerging obesity epidemic in most industrialized societies, additional investigation is needed to determine whether CRC represents another disease entity associated with, or resulting from, IR syndrome.^{46,57} Further development of quantitative IR biomarkers that accurately reflect long-term insulin and glucose exposure might also be rewarding with respect to identifying population subsets that are at increased CRC risk.

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