

Null Results in Brief

IGF-I, IGFBP-3, and IGF-I/IGFBP-3 Ratio: No Association with Incident Colorectal Cancer in the Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study

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Introduction

The insulin-like growth factor (IGF) signaling system is thought to affect tissue growth and development (1), with IGF-I and IGF binding protein-3 (IGFBP-3) having putative procarcinogenic and anticarcinogenic properties, respectively. A recent meta-analysis of four prospective studies found that IGF-I levels were positively associated with incident colorectal cancer (CRC), whereas IGFBP-3 and the IGF-I/IGFBP-3 molar ratio were less clearly associated (2). Two subsequent studies of the IGF proteins and CRC risk showed mixed results (3, 4). Herein, we describe a nested, case-cohort study of fasting serum IGF-I, IGFBP-3, and the IGF-I/IGFBP-3 molar ratio and CRC risk in the Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study.

Materials and Methods

The design and conduct of the Alpha-Tocopherol, Beta-Carotene Study have been reported (5). In brief, 29,133 male cigarette smokers, ages 50 to 69 years, living in southwestern Finland with no previous cancer diagnosis were recruited between 1985 and 1988 and provided fasting serum samples that were archived at -70°C . Demographic, anthropometric, and exposure data were collected using self-reported questionnaires and focused physical examination. Incident cancer cases were identified through the Finnish Cancer Registry, which provides nearly 100% case ascertainment (6).

To facilitate multiple cancer site investigations of serum-based biomarkers, a randomly selected subcohort of 400 Alpha-Tocopherol, Beta-Carotene subjects, alive

and without a cancer diagnosis during the first 5 years of follow-up, was assembled and 134 incident CRC cases diagnosed by December 31, 1997 (range, 5-12 years; median 9 years after serum sampling), were included in this study (one subcohort subject was later excluded due to misclassification of CRC-free status). CRC diagnoses (ICD-9 codes 153.0-153.4, 153.6-153.9, and 154.0-154.1) were confirmed through medical record review by two study physicians. Descriptions of the CRC cases and subcohort controls have been reported (7).

Serum IGF-I and IGFBP-3 levels were determined (M.J. Pollak, Jewish General Hospital and McGill University) by ELISA using reagents from Diagnostic Systems Laboratory. The IGF-I/IGFBP-3 molar ratio was calculated based on 1 ng/mL IGF-I = 0.130 nmol IGF-I and 1 ng/mL IGFBP-3 = 0.036 nmol IGFBP-3.

Spearman partial correlation coefficients examined pairwise associations between IGF serum marker levels among subcohort control subjects, adjusting for age. Relative risks (RR) and 95% confidence intervals (95% CI) were estimated from Cox proportional hazards regression models accounting for the case-cohort study design (8) and applying a robust jackknife variance estimate to correct for the oversampling of cases (9, 10). Survival was modeled as a function of age, which predicted CRC risk better than follow-up time (11). Age-adjusted and multivariable-adjusted RRs were estimated overall and for proximal (ICD-9 codes 153.0, 153.1, 153.4, 153.6, and 153.7) and distal (ICD-9 codes 153.2, 153.3, 154.0, and 154.1) CRC subsites. Selection of covariates for the final multivariate analyses has been described (7). Comparing any two quartiles, assuming a two-sided test of the hypothesis and a type I error rate of 0.05, the study had 80% power to detect quartile-specific RRs as low as 1.99, 2.33, and 3.14 for overall, distal, and proximal CRCs, respectively. All statistical tests were done two-sided, using SAS (SAS Institute, Inc.) and S-Plus (Insightful, Inc.) software.

Results

Median values (interquartile ranges) for IGF-I, IGFBP-3, and IGF-I/IGFBP-3 were 137 (109-165) ng/mL, 2,300

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Table 1. Age-adjusted Spearman correlation between serum IGF markers and other baseline exposures among subcohort control subjects

	IGF-I	IGFBP-3	IGF-I/IGFBP-3	BMI	Alcohol	Physical activity
IGF-I	—	0.71 (<0.001)	0.66 (<0.001)	0.12 (0.02)	-0.08 (0.10)	-0.08 (0.13)
IGFBP-3		—	-0.002 (0.97)	0.18 (<0.001)	-0.01 (0.79)	-0.08 (0.12)
IGF-I/IGFBP-3			—	-0.003 (0.96)	-0.11 (0.03)	-0.05 (0.36)
BMI				—	0.02 (0.68)	-0.04 (0.49)
Alcohol					—	-0.0002 (0.99)
Physical activity*						—

NOTE: Values represent rank-based Spearman correlation coefficient (*P* value), after partialing out the effects of age; *P* values test whether correlation differs from zero.

*Occupational physical activity, coded ordinally as 1, nonworker or sedentary; 2, light; 3, moderate; and 4, heavy.

(1,920-2,753) ng/mL, and 0.21 (0.18-0.25) among CRC cases and 139 (113-175) ng/mL, 2,338 (1,952-2,827) ng/mL, and 0.22 (0.19-0.25) among subcohort controls, respectively. Age-adjusted correlations between the serum biomarkers and other factors are shown in Table 1. The serum-based biomarkers were not significantly associated with incident CRC, overall or by anatomic subsite (Table 2). Body mass index, total energy intake, alcohol intake, and physical activity level did not modify the risk estimates (*P* interaction > 0.05 for each comparison).

Discussion

We found no significant associations between fasting IGF-I, IGFBP-3, or IGF-I/IGFBP-3 levels and CRC risk in this nested case-cohort study, overall or by anatomic subsite. Our findings for IGF-I contrast with data from the only meta-analysis reported to date (pooled odds

ratio, 1.58; 95% CI, 1.11-2.27 comparing extreme quartiles; ref. 2); however, data from subsequent studies have been inconsistent. Wei et al. (3) showed a positive association in a study of 532 women (RR, 2.17; 95% CI, 0.96-4.88 comparing extreme quartiles; *P* trend = 0.03), whereas Otani et al. (4), studying 537 women and 588 men, did not (odds ratio, 0.83; 95% CI, 0.38-1.80; *P* trend = 0.60; and odds ratio, 0.83; 95% CI, 0.40-1.70; *P* trend = 0.91, respectively). With respect to IGFBP-3 and IGF-I/IGFBP-3, we and others (2, 4) have found null associations with CRC risk, whereas Wei and colleagues reported a risk estimate of 2.82 (95% CI, 1.35-5.88) for colon cancer among women in the highest versus lowest quartiles of IGF-I/IGFBP-3 (3).

The strengths of our study include analyses of fasting serum samples obtained at least 5 years prior to incident CRC diagnosis, which minimized the possibility that the observed IGF levels were influenced by physiologic factors or lifestyle changes induced by subclinical

Table 2. Associations between IGF-I, IGFBP-3, IGF-I/IGFBP-3 and incident CRC, overall and by anatomic subsite

	Controls	Any CRC		Proximal CRC		Distal CRC	
		Events	RR (95% CI)*	Events	RR (95% CI)*	Events	RR (95% CI)*
IGF-I (ng/mL)							
Quartile 1 (≤113)	99	36	1.00 (ref.)	13	1.00 (ref.)	23	1.00 (ref.)
Quartile 2 (114-138)	99	33	0.76 (0.41-1.41)	11	0.74 (0.30-1.82)	22	0.80 (0.39-1.66)
Quartile 3 (139-174)	100	36	1.13 (0.62-2.06)	12	1.13 (0.46-2.78)	24	1.20 (0.60-2.42)
Quartile 4 (>174)	101	29	0.92 (0.49-1.70)	11	1.02 (0.41-2.49)	18	0.90 (0.43-1.88)
<i>P</i> trend [†]			0.90		0.78		0.95
IGFBP-3 (ng/mL)							
Quartile 1 (≤1,956)	100	37	1.00 (ref.)	13	1.00 (ref.)	24	1.00 (ref.)
Quartile 2 (1,957-2,334)	99	32	1.01 (0.52-2.00)	10	0.92 (0.30-2.87)	22	1.13 (0.52-2.44)
Quartile 3 (2,335-2,827)	100	36	1.29 (0.69-2.41)	13	1.57 (0.61-3.99)	23	1.22 (0.59-2.49)
Quartile 4 (>2,827)	100	29	0.98 (0.51-1.88)	11	1.58 (0.54-4.63)	18	0.80 (0.38-1.68)
<i>P</i> trend [†]			0.85		0.30		0.67
IGF-I/IGFBP-3							
Quartile 1 (≤0.186)	102	39	1.00 (ref.)	14	1.00 (ref.)	25	1.00 (ref.)
Quartile 2 (0.187-0.217)	98	34	0.95 (0.52-1.71)	12	0.92 (0.38-2.20)	22	0.94 (0.47-1.91)
Quartile 3 (0.218-0.254)	101	37	1.16 (0.65-2.07)	13	1.09 (0.47-2.51)	24	1.23 (0.62-2.43)
Quartile 4 (>0.254)	98	24	0.72 (0.38-1.37)	8	0.58 (0.21-1.63)	16	0.83 (0.40-1.72)
<i>P</i> trend [†]			0.50		0.40		0.84

*IGF-I analyses adjusted for pack-years smoked, body mass index, and fiber intake. IGFBP-3 analyses adjusted for pack-years smoked, body mass index, hypertension, caloric intake, fiber intake, and occupational physical activity. IGF-I/IGFBP-3 ratio analyses adjusted for pack-years smoked, body mass index, and fiber intake. 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 (12). Other variables were included in the final model if any of the following criteria were met: univariately associated with both the exposure and the outcome (*P* < 0.05); inclusion changed the serum biomarker hazard ratio by at least 10%; associated with a *P* ≤ 0.20 in the age-adjusted and smoking-adjusted risk model; or inclusion decreased the standard error for any of the serum biomarker risk estimates.

[†]Based on test for trend.

colorectal neoplasia. Also, the identification of CRC cases and subcohort controls from within the same source population reduced the chance of selection bias. Furthermore, consideration of multiple potential confounding variables, as well as proximal and distal CRC subsites, increased the internal validity of our reported risk estimates.

One potential limitation was our measurement of IGF-I and IGFBP-3 in serum, rather than plasma, samples. As noted by Renehan et al. (2), plasma-based assessments have shown stronger cancer associations than serum-based assessments. The relatively restricted demographic characteristics of our subject population (i.e., older, Finnish male smokers) should also be considered when interpreting the external validity of the current observations. It remains possible that other components of the IGF signaling system, such as IGF-2, may be linked to CRC risk. However, based on the null associations observed with IGF-I and IGFBP-3 in this study, characterization of additional IGF proteins in this case-control set may not be informative.

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