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Plasma Biomarkers of Insulin and the Insulin-like Growth Factor Axis, and Risk of Colorectal Adenoma and Serrated Polyp

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Abstract

Background: Hyperinsulinemia, high insulin-like growth factor 1 (IGF1) levels, and low IGF binding protein 1 (IGFBP1) levels have been implicated in the relationship between obesity and increased risk of colorectal cancer (CRC). However, it remains inconclusive whether circulating biomarkers of insulin and the IGF axis are associated with conventional adenoma and serrated polyp, the two distinct groups of CRC precursors.

Methods: We prospectively examined the associations of plasma C-peptide, IGF1, IGFBP1, IGFBP3, and IGF1 to IGFBP3 ratio with conventional adenoma and serrated polyp among 11 072 women from the Nurses' Health Studies. Multivariable logistic regression was used to calculate the odds ratio (OR) per 1-SD increase in each biomarker for overall risk of conventional adenoma and serrated polyp and according to polyp feature.

Results: During 20 years of follow-up, we documented 1234 conventional adenomas and 914 serrated polyps. After adjusting for various lifestyle factors (including body mass index), higher concentrations of IGFBP1 were associated with lower risk of serrated polyp (OR = 0.84, 95% confidence interval = 0.75 to 0.95, $P = .005$). The association was particularly strong for large serrated polyp (≥ 10 mm) located in the distal colon and rectum (OR = 0.59, 95% confidence interval = 0.39 to 0.87, $P = .01$). In contrast, we did not find any statistically significant association between the biomarkers and conventional adenoma.

Conclusions: A higher plasma level of IGFBP1 was associated with lower risk of serrated polyp. Our findings support a potential role of IGFBP1 in the serrated pathway of CRC in women.

Colorectal cancer (CRC) is the third most common cancer and the third leading cause of cancer death in both men and women in the United States (1). Increasing evidence suggests that CRC represents a group of molecularly heterogeneous diseases that develop through distinct pathways. Although the well-recognized conventional adenoma-carcinoma pathway accounts for approximately 60–80% of CRC (2), serrated polyps contribute another 20–30% of sporadic CRC cases through the

serrated pathway (3). According to the 2010 World Health Organization classification, serrated polyps include hyperplastic polyps (HPs), sessile serrated adenomas or polyps (SSA/Ps), and traditional serrated adenomas (TSAs) (3). The serrated continuum is proposed to mainly originate from HPs and transit to SSA/Ps or TSAs before progression to dysplasia and carcinoma, although some evidence suggests the potential for some SSA/Ps to arise de novo from normal mucosa (4).

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Obesity is an established risk factor for CRC. Increasing data indicate that metabolic disturbances may have a particularly stronger effect on the serrated pathway than the conventional pathway. We recently showed in a large prospective study that obesity was more strongly associated with higher risk of serrated polyp than conventional adenoma (5). A major mechanism underlying the obesity-cancer link is hyperinsulinemia and related changes in the insulin-like growth factor (IGF) axis (6). Both insulin and IGF1 have the potential to directly stimulate the growth and inhibit apoptosis of colonic epithelial and cancer cells (7). Moreover, insulin can downregulate the levels of IGF binding protein-1 (IGFBP1), thereby increasing the levels of bioavailable IGF1 (8). Higher circulating levels of IGF1 and C-peptide (a stable marker of insulin secretion) and lower levels of IGFBP1 have been linked to increased risk of CRC (9). However, evidence is limited on the relationship of prediagnostic insulin-IGF system biomarkers with colorectal premalignant lesions. So far, only a few studies have evaluated C-peptide, IGF1, IGFBP1, and IGFBP3 in relation to conventional adenoma, and the results are conflicting (10–15). Moreover, most of the studies were either cross-sectional or case-control studies with limited sample sizes, making it difficult to disentangle whether the biomarker changes are a cause or consequence of carcinogenesis (16). In addition, to our knowledge, all prior studies have focused on conventional adenoma, and no study has yet examined the biomarker associations with risk of serrated polyp.

Given the ever-increasing epidemic of obesity, a better understanding about the role of metabolic disturbances in early stages of CRC onset is crucial to develop effective prevention strategies to mitigate future CRC risk. Therefore, we examined the associations of prediagnostic C-peptide, IGF1, IGFBP1, and IGFBP3 levels with risk of conventional adenoma and serrated polyp among women in the Nurses' Health Studies (NHS and NHS II).

Methods

Study Population

The NHS and NHS II are two ongoing US cohort studies that included 121 700 registered female nurses aged 30–55 years in 1976 and 116 686 female nurses aged 25–42 years in 1989, respectively. Details about the follow-up of the two cohorts have been described previously (17). In brief, mailed questionnaires were administered biennially to collect updated lifestyle and medical information, with the follow-up rates exceeding 90% for each 2-year cycle in both cohorts. Between 1989 and 1990, 32 826 NHS women donated blood specimens on ice packs by overnight courier; and between 1996 and 1999, 29 611 NHS II women provided blood specimens using a similar method. On receipt, samples were immediately centrifuged, placed in aliquots, and stored in liquid nitrogen freezers (18). Participants who provided blood samples showed similar demographic, dietary, and lifestyle profiles to those who did not (19).

The current study included 10 263 NHS and 4 285 NHS II women with available biomarker data (ie, C-peptide, IGF1, IGFBP1, and IGFBP3) predominately from previous nested case-control studies of various outcomes, including Barrett's esophagus, breast cancer, ovarian cancer, cognitive function (NHS only), colon cancer (NHS only), colon polyps (NHS only), hypertension (NHS only), multiple myeloma (NHS only), myocardial infarction (NHS only), pancreatic cancer (NHS only), type II diabetes (NHS only), benign breast disease (NHS II only), endometriosis (NHS II only), and stroke (NHS II only) (20).

We excluded participants who had biomarker levels considered as outliers by the generalized extreme studentized deviate many-outlier procedure (21); had a history of cancer (except non-melanoma skin cancer), diabetes, cardiovascular disease, colorectal polyp, or inflammatory bowel disease at the time of blood draw; had no lower gastrointestinal endoscopy after blood collection; or had unclassified polyp subtype (see flowchart in [Supplementary Figure 1](#), available online). A total of 11 072 women were included in the final analysis. The study protocol was approved by the institutional review boards of the Brigham and Women's Hospital and Harvard T.H. Chan School of Public Health and those of participating registries as required. Written informed consent was obtained from each participant.

Biomarker Assays

Plasma concentrations of C-peptide were measured by enzyme-linked immunosorbent assay in the laboratory of Dr Michael Pollak or by radioimmunoassay in the laboratories of Dr Robert Cohen and Dr Nader Rifai. IGF1, IGFBP1, and IGFBP3 were measured by enzyme-linked immunosorbent assay in the laboratory of Dr Michael Pollak. The IGF1 to IGFBP3 molar ratio, which has been suggested as an indicator of IGF-I bioavailability, was calculated using the following formula: $\frac{\text{IGF-1 (ng/mL)} \times 0.13}{\text{IGFBP-3 (ng/mL)} \times 0.036}$ (22).

Quality control samples were randomly interspersed among each of the case-control sample sets, and laboratory personnel were blinded to quality control and case-control status for all assays. The lower detection limits and intra-assay coefficient of variation for each biomarker are presented in [Supplementary Table 1](#) (available online). Because biomarkers were measured in multiple batches over time and there might be variation in mean biomarker levels due to differences in reagents, technicians, and laboratories, we recalibrated biomarker concentrations across batches within each cohort to the value of an "average batch" using the method developed by Rosner et al. (23). For each batch, biomarker concentrations (mean [SD]) before and after calibration are presented in [Supplementary Figure 2](#) (available online).

Ascertainment of Colorectal Polyp

Diagnosis of colorectal polyp in the NHS and NHS II has been described in detail previously (5). Briefly, on each biennial questionnaire, participants were asked whether they had undergone a colonoscopy or sigmoidoscopy and whether any colorectal polyp had been diagnosed in the past two years. When a participant reported polyp diagnosis, we asked for permission to obtain her endoscopic and pathologic records, with a success rate of approximately 85%. A study physician, who was blinded to any exposure information, reviewed all records and extracted data on histology, size, number, and anatomic location of polyps. If a participant had more than one polyp in an endoscopy, the histology of the most advanced lesion and the size of the largest polyp were used. The self-report of a negative endoscopy was reliable. In random samples (N = 114) of women who reported having had endoscopy but no polyps, the concordance rate for self-reported negative endoscopy was 97% (24).

In the current study, conventional adenoma included tubular, tubulovillous, and villous adenomas and adenomas with high-grade dysplasia. Advanced conventional adenoma was defined as at least one adenoma greater than or equal to 10 mm in diameter or any size with tubulovillous, villous, or high-grade dysplasia (25). Serrated polyp included HPs and mixed or

serrated adenomas. Mixed or serrated adenoma consisted of both mixed polyps (those with both adenomatous and hyperplastic changes in histology) and polyps with any serrated diagnosis (eg, serrated adenomas, serrated polyps, and SSA/Ps).

Statistical Analysis

The current study included only women who had at least one lower endoscopy since blood draw. If a participant reported more than one endoscopy during the study period, multiple records from the same participant were used in the analysis. Participants were censored at the diagnosis of the first colorectal polyp or the date of last endoscopy, whichever occurred first. To account for multiple records per participant and to handle time-varying covariates efficiently, we used an Andersen-Gill data structure with a new record for each 2-year follow-up period during which a participant underwent an endoscopy.

Our primary hypothesis testing was the associations of C-peptide, IGF1, IGFBP1, IGFBP3, and IGF1 to IGFBP3 ratio with risk of conventional adenoma and serrated polyp. All other analyses, including subgroup analysis according to polyp features, represent secondary analyses. Given the post hoc nature of the study, to account for multiple hypothesis testing, we adjusted the statistical significance level for the 10 primary hypotheses (5 biomarkers \times 2 outcomes) using Bonferroni correction and considered P less than .005 as statistically significant ($\alpha = .05/10 = 0.005$).

We log transformed the concentrations of plasma biomarkers to improve normality. Multivariable logistic regression for clustered data (PROC GENMOD) was used to calculate the odds ratios (ORs) of conventional adenoma and serrated polyp per 1-SD increment in biomarker concentrations. To test for potential nonlinearity, we performed restricted cubic spline analyses and used a likelihood ratio test to compare the model with only the linear term of a biomarker to the model with both the linear and spline terms. We did not find strong statistical evidence for nonlinearity. We also compared the biomarker associations between conventional adenoma and serrated polyp through a case-only analysis and calculated the P for heterogeneity (5). Model 1 was adjusted for age, case or control status in the source case-control studies, fasting status, time period of endoscopy, number of prior endoscopies, time in years since the most recent endoscopy, and study cohort. Model 2 was additionally adjusted for other risk factors for CRC, including race, family history of CRC, height, pack-years of smoking, the Alternate Healthy Eating Index (AHEI) as a measure of diet quality, physical activity (MET-hours/week), alcohol consumption, regular aspirin use, menopausal status, and postmenopausal hormone therapy. Model 3 was further adjusted for body mass index (BMI). P_{trend} was calculated using a Wald test. For continuous covariates, to maximize the ability for confounding control, we calculated the averages based on the two adjacent questionnaires most proximate to blood draw. For missingness (proportions $<2\%$) in selected variables on a questionnaire, we carried forward available information from prior questionnaires.

To evaluate the joint effects of the insulin and IGF1 axis, we cross-classified participants based on the levels of IGFBP1 (median-dichotomized) and other biomarkers (median-dichotomized). $P_{\text{interaction}}$ was assessed using a Wald test for the cross-product terms between the biomarker concentrations (continuous). Subgroup analyses were performed according to histopathological features and anatomical subsites of polyps as well as the median time from blood draw to diagnosis of serrated polyp (<10 and ≥ 10 years). All statistical analyses were

conducted using SAS version 9.4 software (SAS Institute Inc, Cary, NC).

Results

During 20 years of follow-up of 11072 women in the NHS and NHS II, we documented 1234 cases of conventional adenoma and 914 cases of serrated polyp (248 of those also had conventional adenoma). As shown in Table 1, compared with women without any polyp, those with conventional adenomas or serrated polyps were more likely to have a family history of CRC, smoke cigarettes, and drink alcohol and were less likely to regularly use aspirin. C-peptide and IGFBP1 were inversely correlated ($r_s = -0.53$). IGF1 was positively correlated with IGFBP3 ($r_s = 0.57$) and the IGF1 to IGFBP3 molar ratio ($r_s = 0.93$). The other biomarkers were weakly correlated. BMI was positively correlated with C-peptide ($r_s = 0.42$) and inversely with IGFBP1 ($r_s = -0.48$) (Supplementary Table 2, available online).

Table 2 shows the associations between plasma biomarkers and polyp subtypes. Although C-peptide was positively associated with risk of conventional adenoma and serrated polyp in Model 1, further adjustment for covariates, including BMI, attenuated the associations to null (for conventional adenoma: OR per 1 SD = 1.07, 95% confidence interval [CI] = 0.98 to 1.16, $P = .11$; for serrated polyp: OR = 1.08, 95% CI = 0.99 to 1.18, $P = .10$). In the full model that included BMI (Model 3), only IGFBP1 showed an inverse association with risk of serrated polyp at the corrected statistical significance level of 0.005 ($P = .005$), with the OR of 0.84 (95% CI = 0.75 to 0.95) per 1-SD increment in IGFBP1 concentrations. Higher levels of IGF1 and IGFBP3 were associated with increased risk of serrated polyp in the full model, but the associations did not reach statistical significance (OR = 1.09, 95% CI = 1.00 to 1.19, $P = .04$ and OR = 1.09, 95% CI = 1.01 to 1.17, $P = .02$, respectively). In contrast, no association was found between any of the biomarkers and risk of conventional adenoma.

Given the established malignant potential of advanced conventional adenoma and large serrated polyp, we then focused on these high-risk lesions and examined their overall and subsite-specific associations with biomarkers. We combined distal colon and rectal polyps due to the small number of cases. As shown in Table 3, we found that higher IGFBP1 was strongly associated with lower risk of large serrated polyp (OR = 0.63, 95% CI = 0.46 to 0.86, $P = .003$), and the association appeared stronger for large serrated polyp located in the distal colon and rectum than in the proximal colon (OR = 0.59, 95% CI = 0.39 to 0.87, $P = .01$; $P_{\text{heterogeneity}}$ by subsite = .06). C-peptide showed a suggestive positive association with risk of advanced conventional adenomas (OR = 1.13, 95% CI = 1.00 to 1.29, $P = .06$), particularly those located in the proximal colon (OR = 1.27, 95% CI = 1.02 to 1.58, $P = .03$).

To evaluate the joint effect of biomarkers on advanced conventional adenoma and large serrated polyp, we cross-classified individuals based on the levels of IGFBP1 and the other biomarkers and found that women with high IGFBP1 and low IGF1 had the lowest risk of developing large serrated polyp (OR = 0.30, 95% CI = 0.10 to 0.89, $P = .03$ for IGFBP1 above median and IGF1 below median) (Supplementary Figure 3, available online). However, the interaction was not statistically significant ($P_{\text{interaction}} = .16$).

In stratified analysis by time from blood draw to polyp diagnosis (Supplementary Table 3, available online), we did not

Table 1. Basic characteristics of female participants from NHS and NHS II*

Variable	Control group	Conventional adenoma	Serrated polyp
No. of participants	9172	1234	914
Caucasian, %	96	97	97
Age at blood draw, y	53.9 (8.0)	54.9 (7.9)	52.9 (7.8)
Blood fasting, %	76	79	79
No. of endoscopies over follow-up	2.8 (1.8)	2.0 (1.4)	2.1 (1.4)
Time since most recent endoscopy, y	2.6 (2.9)	2.1 (3.2)	2.6 (3.5)
Height, cm	164.3 (6.1)	164.6 (6.0)	164.6 (5.6)
BMI, kg/m ²	25.4 (4.8)	25.6 (4.4)	25.8 (4.7)
Family history of colorectal cancer, %	24	31	26
Pack-year of smoking	9.1 (15.4)	11.4 (16.5)	12.7 (17.3)
Never, %	51	47	41
Past, %	40	40	40
Current, %	9	13	18
Alcohol intake, g/d	5.7 (9.0)	6.3 (9.3)	7.1 (10.2)
Physical activity, MET-hours/wk†	18.7 (19.9)	17.7 (18.1)	19.1 (17.9)
AHEI dietary score	53.1 (10.2)	52.5 (9.6)	52.1 (9.4)
Regular aspirin use, %‡	48	44	44
Postmenopause, %	57	59	56
Current postmenopausal hormone use, %	50	44	42
Biomarker§			
C-peptide, ng/mL	1.52 (1.11–2.22)	1.58 (1.13–2.28)	1.56 (1.19–2.21)
IGF1, ng/mL	159.6 (125.0–200.0)	155.4 (120.0–195.1)	165.8 (128.1–206.5)
IGFBP1, ng/mL	30.1 (15.5–48.5)	26.1 (14.8–44.1)	23.6 (13.2–37.3)
IGFBP3, ng/mL	4317 (3786–4934)	4334 (3821–4926)	4375 (3885–4967)
IGF1 to IGFBP3 molar ratio	0.14 (0.11–0.16)	0.13 (0.10–0.16)	0.14 (0.11–0.16)

*All variables are adjusted for age except for age itself. Mean (SD) is presented for continuous variables and percentage for categorical variables, unless otherwise specified. AHEI, Alternative Healthy Eating Index; BMI = body mass index; IGF1, insulin-like growth factor 1; IGFBP1, insulin-like growth factor binding protein 1; IGFBP3, insulin-like growth factor binding protein 3; MET, metabolic equivalent task; NHS = Nurses' Health Study.

†Physical activity is represented by the product sum of the METs of each specific recreational activity and hours spent on that activity per week.

‡A standard tablet contains 325 mg aspirin, and regular users were defined as those who used at least two tablets per week.

§The natural-log transformed biomarker concentrations were back transformed and presented as median values (quartiles).

|| $(\text{IGF1 [ng/mL]} \times 0.13)/(\text{IGFBP3 [ng/mL]} \times 0.036)$.

observe any statistically significant heterogeneity in the associations across the subgroups.

We also performed a sensitivity analysis by restricting to fasting samples only. As shown in [Supplementary Table 4](#) (available online), the results were similar to our primary findings.

Discussion

In this prospective analysis of 11 072 women in the NHS and NHS II cohorts, we found that higher levels of IGFBP1 were associated with lower risk of serrated polyp, particularly large serrated polyp located in the distal colon and rectum. No statistically significant association was found between any of the biomarkers and risk of conventional adenoma.

Obesity and weight gain have been linked to lower circulating levels of IGFBP1 (20,26,27) but have little effect on other components of the IGF axis, such as IGF1 and IGFBP3 in men and women (28,29). IGFBP1 is primarily secreted by hepatocytes and negatively regulated by insulin. Therefore, plasma levels of IGFBP1 are thought to robustly reflect end organ stimulation by insulin and proposed as a specific marker for hepatic insulin sensitivity (30). Prospective studies have associated lower baseline IGFBP1 levels with increased risk of CRC (31,32). Mechanistically, because the affinity of IGFBP1 for IGF1 exceeds that of IGF1 for the type-1 IGF-receptor, high IGFBP1 levels may reduce IGF1 bioavailability and thus inhibit the insulin-like activity of IGF1 on peripheral metabolism (33). Additionally, in

contrast to the stabilizing effect of IGFBP3 on IGF1, IGFBP1 is involved in modulating the acute bioavailability of IGFs (34). Biological evidence also suggests that IGFBP1 may exert inhibitory effects on cancer cell growth through IGF-dependent and independent pathways (35,36). Therefore, these data are in line with our current findings and together support a potential role of IGFBP1 in the relationship between obesity and serrated polyp.

Of note, we observed a stronger association between IGFBP1 and large serrated polyp (≥ 10 mm), which has been associated with higher risk of CRC incidence (37,38). Given the challenges in endoscopic detection of serrated polyps and malignant potential of large serrated polyps, our findings suggest the potential utility of IGFBP1 in targeted colonoscopic screening for serrated polyps. Moreover, our stratified analysis by subsite showed a stronger association for large serrated polyps located in the distal colon and rectum than those in the proximal colon. This finding is consistent with previous data that showed a stronger association between obesity and serrated polyp in the distal colon and rectum than in the proximal colon (5,39). However, considering the limited number of cases by subgroups of serrated polyps in the current study, future investigations are needed to confirm our findings and uncover the underlying mechanisms for IGFBP1 in the serrated pathway.

The relationship between circulating C-peptide levels and CRC risk has been well studied, with a positive relationship consistently observed (40). However, data on the association of C-peptide with CRC precursors, including conventional adenoma and serrated polyp, remain inconclusive. Several studies have

Table 2. Associations of plasma biomarkers for the insulin-IGF system with conventional adenoma and serrated polyp in women from NHS and NHS II*

Biomarker	Conventional adenoma			Serrated polyp			P _{heterogeneity}
	No.	OR (95% CI) per 1 SD	P	No.	OR (95% CI) per 1 SD	P	
C-peptide							
Model 1†	897	1.11 (1.03 to 1.19)	.004	584	1.14 (1.05 to 1.24)	.002	.50
Model 2‡	897	1.10 (1.02 to 1.18)	.02	584	1.12 (1.03 to 1.22)	.01	.63
Model 3§	897	1.07 (0.98 to 1.16)	.11	584	1.08 (0.99 to 1.18)	.10	.53
IGF1							
Model 1†	1031	1.01 (0.95 to 1.07)	.81	721	1.07 (0.99 to 1.16)	.10	.17
Model 2‡	1031	1.00 (0.93 to 1.07)	.95	721	1.07 (0.98 to 1.16)	.12	.18
Model 3§	1031	1.01 (0.95 to 1.08)	.78	721	1.09 (1.00 to 1.19)	.04	.15
IGFBP1							
Model 1†	699	0.90 (0.83 to 0.97)	.01	384	0.80 (0.72 to 0.88)	<.0001	.03
Model 2‡	699	0.93 (0.85 to 1.01)	.07	384	0.80 (0.72 to 0.89)	<.0001	.02
Model 3§	699	0.97 (0.88 to 1.06)	.47	384	0.84 (0.75 to 0.95)	.005	.045
IGFBP3							
Model 1†	1078	1.05 (0.99 to 1.11)	.12	766	1.10 (1.02 to 1.18)	.01	.38
Model 2‡	1078	1.04 (0.98 to 1.11)	.15	766	1.09 (1.01 to 1.17)	.02	.52
Model 3§	1078	1.04 (0.98 to 1.10)	.17	766	1.09 (1.01 to 1.17)	.02	.50
IGF1 to IGFBP3							
Model 1†	1031	0.96 (0.90 to 1.03)	.27	721	1.00 (0.93 to 1.09)	.92	.25
Model 2‡	1031	0.95 (0.89 to 1.02)	.16	721	1.00 (0.92 to 1.09)	1.00	.21
Model 3§	1031	0.97 (0.91 to 1.04)	.38	721	1.03 (0.95 to 1.12)	.50	.17

*SDs for C-peptide, IGF1, IGFBP1, IGFBP3, IGF1 to IGFBP3 were 1.18, 57.84, 26.49, 906.35, and 3.91, respectively. BMI = body mass index; CI = confidence interval; IGF1 = insulin-like growth factor 1; IGFBP1 = insulin-like growth factor binding protein 1; IGFBP3 = insulin-like growth factor binding protein 3; NHS = Nurses' Health Study; OR = odds ratio.

†Adjusted for age, case or control status, fasting status, time period of endoscopy, number of prior endoscopies, and time in years since the most recent endoscopy.

‡Additionally adjusted for race, family history of colorectal cancer, height, pack-years of smoking, Alternative Healthy Eating Index score, physical activity, alcohol consumption, regular aspirin use, menopausal status, and hormone therapy.

§Further adjusted for BMI. P_{heterogeneity} was calculated through case-only analysis by comparing serrated polyp with conventional adenoma.

investigated the association between circulating C-peptide and risk of conventional adenoma and mostly reported a positive association in men (10,15,41). However, most of the studies are case-control studies and vulnerable to reverse causality, because blood samples were collected from cases after adenoma development. Three prospective studies have evaluated the relationship between C-peptide levels and conventional adenoma but reported mixed results, with one reporting a positive association in women (42) and the others a null result in both sexes (12,43). In the current study, we observed no significant association between C-peptide and conventional adenoma or serrated polyp in women, although those with higher levels of C-peptide showed a borderline significantly elevated risk of advanced conventional adenoma or large serrated polyp. These findings suggest that insulin might primarily act as a promoter rather than initiator in the conventional and serrated pathways.

Some studies have evaluated the associations of IGF1 and IGFBP3 with conventional adenoma, but the results are inconsistent. For example, one cross-sectional study including men and women reported a positive association between IGF1, IGF1 to IGFBP3 ratio, and the presence of adenoma (13), whereas some case-control studies did not replicate the association (11,14,15). In a meta-analysis, levels of IGF1, IGFBP3, and IGF1 to IGFBP3 ratio were not associated with conventional adenoma, but IGF1 was positively associated with advanced adenoma (16). Consistent with most studies included in the meta-analysis, we found no statistically significant associations of plasma IGF1, IGFBP3, and IGF1 to IGFBP3 ratio with conventional adenoma. Given an established, albeit modest, relationship between IGF1 and CRC (44,45), it is possible that the role of the IGF axis

primarily occurs in the later stage of the adenoma-to-carcinoma sequence. Moreover, as in previous studies, we only measured concentrations of total IGF1 rather than free forms that exert bioactive effects. Although widely used, the validity of the IGF1 to IGFBP3 molar ratio as an indicator of IGF1 bioavailability has yet to be experimentally confirmed (34). Therefore, our null findings for total IGF1 cannot rule out the possibility that free IGF1 may have an effect on the development of CRC precursor lesions.

In our study, elevated levels of IGFBP3 were nominally associated with higher risk of total serrated polyp but not with large serrated polyp. A prospective cohort study reported that IGFBP3 was inversely associated with colon cancer risk in men (46), whereas other cohort studies found a positive (47) or null association (44). Further investigation is necessary to clarify the role of IGFBP3 in colorectal carcinogenesis.

The major strengths of this study include large sample size, prediagnostic blood draw, long-term follow-up, and detailed data on covariates that allow for robust confounding control. Moreover, we performed systematic ascertainment and review of different subtypes of colorectal polyps with detailed collection of histopathological data and were thus able to directly compare conventional adenoma and serrated polyp and examine the associations according to polyp features. The study also has several limitations. First, due to the evolving nature and lack of consensus on the diagnostic criteria for specific subtypes of serrated polyps, we were unable to separate HPs, SSA/Ps, and TSAs based on the review of pathology records. However, as indicated by prior data and proposed by an expert panel, serrated polyps larger than 10 mm are a good indicator for SSA/P (48).

Table 3. Association of plasma biomarkers for the insulin-IGF system with advanced conventional adenoma and large serrated polyp in women from NHS and NHS II*

Biomarker	Advanced conventional adenoma			Large serrated polyp (≥10 mm)		
	Overall	Proximal colon	Distal colon and rectum	Overall	Proximal colon	Distal colon and rectum
C-peptide						
No.	337	116	273	62	31	40
OR (95% CI) per 1 SD	1.13 (1.00 to 1.29)	1.27 (1.02 to 1.58)	1.08 (0.94 to 1.24)	1.22 (0.94 to 1.58)	1.19 (0.85 to 1.66)	1.16 (0.82 to 1.64)
P _{trend}	.06	.03	.30	.14	.30	.41
P _{heterogeneity by subsite}			.12			.78
IGF1						
No.	375	126	306	70	38	42
OR (95% CI) per 1 SD	0.95 (0.85 to 1.06)	0.87 (0.72 to 1.04)	0.99 (0.88 to 1.11)	0.96 (0.77 to 1.20)	0.96 (0.67 to 1.37)	1.05 (0.82 to 1.35)
P _{trend}	.34	.12	.85	.74	.81	.69
P _{heterogeneity by subsite}			.16			.55
IGFBP1						
No.	276	82	238	35	16	24
OR (95% CI) per 1 SD	0.95 (0.82 to 1.10)	0.84 (0.64 to 1.11)	0.97 (0.82 to 1.14)	0.63 (0.46 to 0.86)	0.85 (0.53 to 1.36)	0.59 (0.39 to 0.87)
P _{trend}	.50	.22	.68	.003	.50	.01
P _{heterogeneity by subsite}			1.00			.06
IGFBP3						
No.	388	134	314	74	40	45
OR (95% CI) per 1 SD	0.99 (0.90 to 1.09)	0.98 (0.83 to 1.15)	0.99 (0.90 to 1.10)	0.94 (0.76 to 1.18)	0.93 (0.69 to 1.27)	0.98 (0.76 to 1.26)
P _{trend}	.83	.77	.92	.60	.66	.86
P _{heterogeneity by subsite}			.42			.25
IGF1 to IGFBP3						
No.	375	126	306	70	38	42
OR (95% CI) per 1 SD	0.95 (0.85 to 1.06)	0.87 (0.73 to 1.04)	0.99 (0.88 to 1.12)	1.00 (0.78 to 1.29)	1.00 (0.66 to 1.51)	1.08 (0.84 to 1.38)
P _{trend}	.35	.12	.91	1.00	1.00	.56
P _{heterogeneity by subsite}			.04			.54

*Multivariate models were adjusted for age, case or control status, fasting status, time period of endoscopy, number of prior endoscopies, time in years since the most recent endoscopy, race, family history of colorectal cancer, height, pack-years of smoking, AHEI score, body mass index, physical activity, alcohol consumption, regular aspirin use, menopausal status, and hormone therapy. P_{heterogeneity} was calculated through case-only analysis (distal colon and rectum vs proximal colon). AHEI = Alternate Healthy Eating Index; CI = confidence interval; IGF1 = insulin-like growth factor 1; IGFBP1 = insulin-like growth factor binding protein 1; IGFBP3 = insulin-like growth factor binding protein 3; NHS = Nurses' Health Study; OR = odds ratio.

Second, as is typical of previous studies, a single biomarker measurement was available for each participant and may not reflect long-term exposures. However, existing evidence indicates that levels of the biomarkers included in the study are generally stable over time (49–51). Third, our study participants were female health professionals of predominately European ancestry, which limits the generalizability of the findings. However, no clear evidence indicates that the biological role of the insulin-IGF system in CRC varies by sex or populations. On the other hand, the homogeneity of the study population minimized the likelihood of residual confounding.

In summary, we found that higher IGFBP1 levels were associated with lower risk of serrated polyp, particularly large serrated polyp located in the distal colon and rectum. Our findings indicate a potential role of IGFBP1 in the serrated pathway for colorectal carcinogenesis. Further studies are needed to confirm our findings and elucidate the underlying mechanisms.

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