COLORECTAL CANCER

# Plasma insulin-like growth factor 1, insulin-like growth factor binding protein 3, and risk of colorectal cancer: a prospective study in northern Sweden

**R Palmqvist, G Hallmans, S Rinaldi, C Biessy, R Stenling, E Riboli, R Kaaks**

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**Background:** Insulin-like growth factor 1 (IGF-1) has antiapoptotic and mitogenic effects on various cell types, and raised IGF-1 levels are increasingly being implicated as potential risk factors for cancer.
**Aims:** To examine the relationship between IGF-1 and its major plasma binding protein, IGF binding protein 3 (IGFBP-3), and the risk of colorectal cancer.
**Methods:** We conducted a case-control study nested within the Northern Sweden Health and Disease Cohort. IGF-1 and IGFBP-3 were measured in prediagnostic plasma samples from 168 men and women who developed cancers of the colon (n=110) or rectum (n=58), and from 336 matched controls.
**Results:** Conditional logistic regression analyses showed an increase in colon cancer risk with increasing levels of IGF-1 (odds ratios (ORs) 1.00, 1.89, 2.30, 2.66; $p_{trend}$=0.03) and IGFBP-3 (ORs 1.00, 0.91, 1.80, 1.93; $p_{trend}$=0.02). Rectal cancer risk was inversely related to levels of IGF-1 (ORs 1.00, 0.45, 0.33, 0.33; $p_{trend}$=0.09) and IGFBP-3 (ORs 1.00, 0.75, 0.66, 0.49; $p_{trend}$=0.21). Mutual adjustments between IGF-1 and IGFBP-3 did not materially alter these relationships.
**Conclusions:** These results support earlier findings of increased risk of colon cancer in subjects with elevated plasma IGF-1. Our results however do not support the hypothesis that the risk of rectal cancer could also be directly related to IGF-1 levels.

See end of article for authors' affiliations

Correspondence to: R Kaaks, International Agency for Research on Cancer, 150 Cours Albert Thomas, 69372 Lyon, Cedex 08, France; kaaks@iarc.fr

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Insulin-like growth factor 1 (IGF-1)—a strongly mitogenic and antiapoptotic factor—is being increasingly implicated in the development of human cancers, including cancer of the colorectum.[1–3] The IGF-1 receptor (IGF1-R) is expressed in normal non-transformed colonic mucosa, as well as in human colorectal cancers.[3 4] Stimulation by IGF-1 causes proliferation in vitro while blocking IGF1-R inhibits growth of colorectal cancer cells.[5 6] Cells from mice homozygous for a disrupted IGF1-R gene are refractory to transformation by oncogenes.[7] In humans, acromegaly, a medical condition with strongly elevated plasma IGF-1 levels, is associated with an increased proliferation of normal colonic epithelium[8] and with an increased risk of developing colorectal adenomas and cancer.[9–12]

The bioactivity of IGF-1 within tissues depends on circulating levels and local production of IGF-1, and of at least six different IGF binding proteins (IGFBPs).[13] In the circulation, IGFBPs regulate the efflux of IGF-1 towards target tissues, and at the tissue level they modulate IGF-1 bioactivity by inhibiting or enhancing binding of IGF-1 to its receptors. The major plasma IGF binding protein is IGFBP-3 which binds more than 90% of IGF-1. The main stimulatory factor for synthesis of IGF-1 and IGFBP-3 is growth hormone (GH).

Levels of total circulating IGF-1, as well as IGFBPs 1, 2, and 3, are influenced by variations in nutritional status, particularly energy balance.[14 15] Chronic energy restriction, a strongly protective factor against various forms of cancer, including the colorectum, strongly reduces plasma levels of IGF-1 and IGFBP-3, and increases levels of IGFBP-1 and IGFBP-2. On the other hand, positive energy balance and obesity cause hyperinsulinaemia and lead to reductions in plasma IGFBP-1 and IGFBP-2.

Several prospective cohort studies have recently provided evidence for an increase in colorectal cancer risk in men and women with elevated circulating IGF-1, as total concentration or relative to IGFBP-3.[16–18] In this report, we present results from a cohort of men and women in the Northern Sweden Health and Disease Study, relating the risk of colon cancer and rectal cancer to plasma concentrations of IGF-1 and IGFBP-3.

## SUBJECTS AND METHODS

### Study cohort

Subjects were recruited to the Northern Sweden Health and Disease cohort study through the Västerbotten Intervention Project (VIP), the Northern Sweden part of the WHO study for Monitoring of Trends and Cardiovascular Disease Study (MONICA), and an ongoing Mammary Screening Project (MSP). VIP started as a population based intervention study with the aim of decreasing mortality due to cardiovascular disease by advocating a healthy diet and lifestyle to the general public. In this project, which started in 1986 and which is still recruiting new subjects, all residents in the county of Västerbotten (total population 260 000) are invited to a health survey in the years in which they become 30, 40, 50, or 60 years old. In March 2000, a total of 30 300 men and 31 900 women, aged 30–62 at recruitment, were included. The MONICA study includes 5374 men and 5500 women, recruited in 1986, 1990, 1994, and 1999, as a representative population sample from the counties of Västerbotten and Norrbotten, and who were between 41 and 70 years of age. MSP started in 1995, and in March 2000 included 32 800 women, aged 50–70 years, who were all screened at least once. MSP is currently still recruiting new subjects.

**Abbreviations:** IGF-1, insulin-like growth factor 1; IGF1-R, IGF-1 receptor; IGFBP-3, IGF binding protein 3; GH, growth hormone; VIP, Västerbotten Intervention Project; MONICA, Monitoring of Trends and Cardiovascular Disease Study; MSP, Mammary Screening Project; BMI, body mass index; OR, odds ratio; IRMA, immunoradiometric assay.

**Table 1** Anthropometry indices, smoking status, and mean serum hormone measurements in cases with colorectal cancer and control subjects

| | Controls | Colorectal cancer | p for difference |
|---|---|---|---|
| No of individuals | 336 | 168 | |
| Height (m) | 169.4 (9.3) | 169.7 (8.3) | 0.76 |
| Weight (kg) | 74.7 (13.5) | 76.0 (13.4) | 0.32 |
| BMI (kg/m$^2$) | 26.0 (3.8) | 26.3 (3.8) | 0.50 |
| Current smokers (%) | 20.5 | 16.7 | 0.30 |
| IGF-1 (ng/ml) | 200.4 [192.6–208.2] | 198.7 [188.1–209.3] | 0.78 |
| IGFBP-3 (ng/ml) | 2585 [2504–2666] | 2593 [2490–2696] | 0.91 |

Values are mean (population standard deviations) or mean [95% confidence intervals (CI)]; CI=mean (1.96)*(stdev/sqrt(n)).
BMI, body mass index; IGF-I, insulin-like growth factor 1; IGFBP-3, IGF binding protein 3.

In each of these projects, subjects were asked to complete a self administered questionnaire to obtain demographic, medical, and lifestyle information. The lifestyle questionnaires included questions on smoking status (current smoker, exsmoker, non-smoker) and diet. In addition, anthropometric measurements (height, weight) were recorded, and a 20 ml blood sample was obtained. Weight was measured without shoes and in light indoor clothing, and was recorded to the closest 0.2 kg. Height was measured without shoes, and recorded to the closest centimetre. In all three subcohorts, blood was collected in one heparin tube and one EDTA tube, centrifuged, aliquoted as plasma and buffy coat, frozen at −20°C or −80°C, and transferred within one week to a −80°C central storage facility (the Northern Sweden Medical Research Biobank). The procedures for blood donation, processing, and storage as aliquots of plasma and buffy coat were the same in all three subcohorts, except for fasting/non-fasting status. Blood samples from the majority of study subjects, who had been recruited through the VIP and MONICA projects, were obtained in the morning, and fasting time before blood donation was more than eight hours in 55%, 4–8 hours in 32%, and less than four hours in 13%. In MSP however none of the women had been asked to provide blood samples under fasting conditions, and 98% had fasted for less than four hours before blood donation. All participants gave written consent for use of their blood samples in future research projects, and the study was approved by the ethics committee of Umeå University.

All incident cases of colorectal cancer and all deaths were identified through linkage with regional and national cancer registries, and with regional and national registries for all-cause mortality. Linkage was done using a national individual identification number as the identity link, and with a high degree of completeness of case ascertainment. We identified 168 incident cases of colorectal cancer, diagnosed after blood donation, up to March 1999. All cancer cases were verified histologically by reviewing medical records. Six cases were excluded because their pathology records did not confirm the cancer diagnosis indicated by the cancer registries. Two further cases were excluded because their plasma samples had been used for other research projects. We thus retained 168 colorectal cancer cases for the study. Of the 168 cancer cases retained for the study, 93 were women and 75 were men; of these, 113 had been recruited in the VIP, 41 cases in the MSP, and 14 in the MONICA subcohorts. The subsite distribution of the 168 colorectal cancers was as follows: 58 in the right colon (caecum, ascending colon and hepatic flexure, transverse colon), 52 in the left colon (splenic flexure, descending colon, and sigmoid colon), and 58 in the rectum (rectosigmoid junction, rectum). Twenty three cases were classified by the pathologist as Dukes' stage A, 58 as Dukes' B, and 55 as Dukes' C, and 32 cases had unspecified Dukes' stage.

Controls were selected randomly from all cohort members alive and free of cancer at the time of diagnosis of a given index case, and by matching the case for sex, subcohort, age (±6 months) and date (±2 months) at blood sampling, and fasting time (<4 hours, 4–8 hours, or >8 hours).

Time intervals between blood donation and cancer diagnosis ranged from less than 1.5 months up to 10 years (median 3.35 years). Of the 168 cases, 146 (87%) were diagnosed more than one year, 118 (70%) more than two years, and 68 (40%) more than four years after blood donation.

## Laboratory analyses

Stored, never thawed plasma samples (from originally heparinised blood samples) of the cancer cases and their matched controls were identified, packed on dry ice, and shipped to the International Agency for Research on Cancer (Lyon, France) for hormone measurements. IGF-1 and IGFBP-3 were measured by double antibody immunoradiometric assays (IRMA) using reagents from Immunotech (Marseille, France). The protocol for the IGF-1 assay included an acid-ethanol extraction step to release IGF-1 from its binding proteins. The laboratory personnel were blinded as to the case-control status of the plasma samples. Samples pertaining to matched study subjects were always analysed together in the same batch (that is, on the same day and within the same immunoassay kit). To control the quality of each type of peptide measurement, analytical batches systematically included three standard sera. The average intra-batch coefficients of variation calculated from these standard sera were 10.9% and 4.5%, respectively, for IGF-1 and IGFBP-3.

## Statistical analyses

Pearson's coefficients of correlation adjusted for age, sex, and case-control status were used to examine the cross sectional relationships between IGF-1, IGFBP-3, height, and body mass index (BMI=height/weight$^2$).

A pairwise *t* test was used to test for mean differences between anthropometric indices or hormone levels of the cases, and the average value for the two matched controls. Odds ratios (ORs) for disease were calculated by conditional logistic regression for quartile levels of IGF-1 and IGFBP-3. Quartile cut off points were determined on variable distributions of cases and controls combined. Confidence intervals (95%) were computed using the standard errors of the pertinent regression coefficients, and assuming a normal probability distribution for the estimated coefficients. Likelihood ratio tests for linear trends in risk with increasing peptide concentrations were performed using scores 1, 2, 3, and 4 for the four quartile levels. All statistical tests and corresponding p values were two sided.

Multivariate logistic regression was used to estimate ORs adjusted for possible confounding factors other than those controlled for by matching. Potential confounding factors included smoking status at the time of blood donation, BMI, and height. In addition, associations of risk with levels of IGF-1 were estimated with adjustment for levels of IGFBP-3. All logistic regression analyses were performed using the "PHREG" procedure for proportional hazards regression of the Statistical Analysis System (SAS).[19]

**Table 2** Odds ratios of cancer of the colorectum, colon, or rectum for quartiles of serum IGF-1 and IGFBP-3

| | Quartile level | | | | |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | p for trend |
| IGF-1 | | | | | |
| Colorectum | | | | | |
| OR | 1.00 | 1.17 | 1.19 | 1.27 | 0.51 |
| 95% CI | — | 0.66–2.08 | 0.63–2.22 | 0.65–2.47 | |
| No of cases/controls | 39/86 | 43/83 | 45/81 | 41/85 | |
| Mean exposure | 109.86 | 172.71 | 222.79 | 293.14 | |
| Colon | | | | | |
| OR | 1.00 | 1.89 | 2.30 | 2.66 | 0.03 |
| 95% CI | — | 0.89–4.04 | 1.02–5.17 | 1.09–6.50 | |
| No of cases/controls | 21/64 | 30/57 | 30/46 | 29/53 | |
| Mean exposure | 105.92 | 172.71 | 222.59 | 293.54 | |
| Rectum | | | | | |
| OR | 1.00 | 0.45 | 0.33 | 0.33 | 0.09 |
| 95% CI | — | 0.13–1.53 | 0.09–1.14 | 0.09–1.13 | |
| No of cases/controls | 18/22 | 13/26 | 15/35 | 12/32 | |
| Mean exposure | 118.24 | 172.72 | 223.11 | 292.41 | |
| IGFBP-3 | | | | | |
| Colorectum | | | | | |
| OR | 1.00 | 0.80 | 1.25 | 1.23 | 0.24 |
| 95% CI | — | 0.44–1.45 | 0.68–2.29 | 0.68–2.22 | |
| No of cases/controls | 40/85 | 35/91 | 46/79 | 46/80 | |
| Mean exposure | 1754 | 2394 | 2740 | 3456 | |
| Colon | | | | | |
| OR | 1.00 | 0.91 | 1.80 | 1.93 | 0.02 |
| 95% CI | — | 0.42–1.94 | 0.82–3.98 | 0.92–4.06 | |
| No of cases/controls | 23/55 | 23/64 | 30/50 | 34/51 | |
| Mean exposure | 1761 | 2405 | 2738 | 3457 | |
| Rectum | | | | | |
| OR | 1.00 | 0.75 | 0.66 | 0.49 | 0.21 |
| 95% CI | — | 0.27–2.06 | 0.23–1.87 | 0.16–1.49 | |
| No of cases/controls | 17/30 | 12/27 | 16/29 | 12/29 | |
| Mean exposure | 1743 | 2370 | 2743 | 3459 | |

Conditional logistic regression analyses were matched for age, sex, date of blood sampling, and fasting status, and further adjusted for smoking status.
OR, odds ratio; CI, confidence interval; IGF-I, insulin-like growth factor 1; IGFBP, IGF binding protein.
Colorectum: IGF-1 measurements were missing for one control; IGFBP-3 measurements were missing for one case and one control.
Rectum: IGF-1 measurements were missing for one control; IGFBP-3 measurements were missing for one case and one control.
Cut off points: IGF-1, 146.35–197.59–252.64; IGFBP-3, 2176–2575–2948.

## RESULTS

Baseline characteristics of cases and controls are shown in table 1. Colon cancers tended to be more advanced (nine Dukes' A, 44 Dukes' B, 39 Dukes' C, 18 unspecified) than cancers of the rectum (14 Dukes' A, 14 Dukes' B, 16, Dukes' C, 14 unspecified), with an almost fourfold higher probability for colon cancers compared with rectal cancers to be Dukes' stage C instead of stage A (OR 3.8). Mean values for height, weight, BMI, and the two peptides did not differ significantly between cases (colon and rectum combined) and controls, and there was there no significant difference in the percentage of current smokers. Nevertheless, cases had a significantly larger percentage of ex-smokers than controls (OR 1.70; $p_{trend}$=0.048). Cases of rectal cancer had a significantly lower mean IGF-1 level (192.3 (174.2–210.4)) than their matched controls (211.6 (199.7–223.5)) while values for colon cancer cases (202 (189.0–215.1)) were slightly higher than their respective controls (194.5 (184.5–204.6)). BMI had a moderate but non-significant direct association with the risk of colon cancer (ORs 1.00, 1.43, 1.32, 1.53; $p_{trend}$=0.33) but not of rectal cancer (ORs 1.00, 0.43, 0.61, 0.85; $p_{trend}$=0.97).

Combining the data for cases and controls, but adjusting for age, sex, and case-control status, we observed positive correlations between IGF-1 and IGFBP-3 ($r$=0.41, p<0.001) but no significant associations for IGF-1 or IGFBP-3 with either height ($r$=0.11, p=0.01) or BMI ($r$=0.06, p=0.19). There were no significant associations for IGF-1 or IGFBP-3 levels with smoking status.

Logistic regression analyses for colon and rectal cancers combined, and adjusting for smoking status at the time of blood donation, showed no significant associations between risk and levels of IGF-1 or IGFBP-3 (table 2). Nevertheless, when the analyses were performed separated for the colon and rectum, the risk of colon cancer was found to be positively and significantly related to plasma levels of IGF-1 and IGFBP-3 whereas the risk of rectal cancer was found to be inversely related to levels of these two peptides (although this was not significant). When we divided the analyses for tumours with Dukes' stages A, B, and C (colon and rectum combined), none of the tumour subcategories showed any risk association with IGF-1 levels (data not shown).

The associations of IGF-1 and IGFBP-3 with risk of colonic and rectal cancers were unaffected by adjustments for BMI or height. Mutual adjustments between IGF-1 and IGFBP-3 also did not alter the associations (table 3). Finally, the associations also remained unaffected when analyses were restricted to cancer cases diagnosed more than one year after blood donation.

## DISCUSSION

In this prospective cohort study, we observed an increase in the risk of colon cancer but a decrease in the risk of rectal cancer with rising levels of circulating IGF-1 and IGFBP-3.

**Table 3** Odds ratios of cancer of the colorectum, colon, or rectum for quartiles of serum IGF-1 and IGFBP-3, mutually adjusted

| | Quartile | | | | |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | p for trend |
| IGF-1 adjusted for IGFBP-3 | | | | | |
| Colorectum | | | | | |
| OR | 1.00 | 1.18 | 1.18 | 1.27 | 0.56 |
| 95% CI | | 0.65–2.14 | 0.60–2.29 | 0.62–2.63 | |
| Colon | | | | | |
| OR | 1.00 | 1.81 | 2.16 | 2.47 | 0.08 |
| 95% CI | | 0.83–3.98 | 0.91–5.14 | 0.93–6.53 | |
| Rectum | | | | | |
| OR | 1.00 | 0.50 | 0.38 | 0.43 | 0.23 |
| 95% CI | | 0.15–1.69 | 0.10–1.41 | 0.11–1.59 | |
| IGFBP-3 adjusted for IGF-1 | | | | | |
| Colorectum | | | | | |
| OR | 1.00 | 0.82 | 1.31 | 1.32 | 0.20 |
| 95% CI | | 0.44–1.54 | 0.68–2.54 | 0.66–2.67 | |
| Colon | | | | | |
| OR | 1.00 | 0.86 | 1.70 | 1.75 | 0.07 |
| 95% CI | | 0.39–1.91 | 0.73–3.94 | 0.72–4.22 | |
| Rectum | | | | | |
| OR | 1.00 | 0.79 | 0.81 | 0.58 | 0.45 |
| 95% CI | | 0.27–2.30 | 0.25–2.66 | 0.16–2.16 | |

Conditional logistic regression analyses were matched for age, sex, date of blood sampling, fasting status, and time of last food consumption and further adjusted for smoking status.
IGF-I, insulin-like growth factor 1; IGFBP, IGF binding protein; OR, odds ratio; CI, confidence interval.

One of the strengths of the prospective design was that for the majority of cases insulin, IGF-1, and IGFBP-3 were measured in blood samples collected well before cancer diagnosis. Thus as none of the cases was aware of their disease at the time of blood donation, blood peptide levels could not have been affected by metabolic alterations related to psychological stress or cancer treatment. Furthermore, the prospective design also makes it unlikely that case-control differences in peptide levels were a consequence, rather than a possible cause, of tumour development. The latter is corroborated by the fact that associations between cancer risk and peptide levels remained unaltered when cases with tumours diagnosed within less than one year from blood donation were excluded from the statistical analyses.

Our finding of an increased risk of colon cancer with increasing levels of circulating IGF-1 concurs with observations in one small case control study,[20] and in two previous cohort studies, one in US male physicians[16] and one in US nurses.[17] In each of these studies however the increase in risk was reported for cancers of the colon and rectum combined, and in the two previous cohort studies risk was significantly related to IGF-1 levels only after adjustment for IGFBP-3. The latter findings led to the hypothesis that, more specifically, risk may be determined more by a relative increase in IGF-1 compared with levels of IGFBP-3 than by elevation in absolute IGF-1 concentrations. This hypothesis was partially founded on the observation that IGFBP-3 can stimulate apoptosis of various cell types in vitro, most probably through a specific IGFBP-3 receptor,[21 22 23] and on the knowledge that one of the major functions of IGFBP-3 in the circulation and within tissues is to decrease the amounts of IGF-1 available for binding to IGF receptors.[13] In the present study however we found that both IGF-1 and IGFBP-3 were positively associated with the risk of colon cancer and adjustment for IGFBP-3 did not strengthen the association of IGF-1 with colon cancer risk. Similar findings were obtained recently in another prospective study—the New York University Womens' Health Study—where adjustment for IGFBP-3 also did not strengthen the association of disease risk with serum IGF-1.[18]

One hypothesis that might explain these contrasting observations on the association of risk with IGFBP-3 and IGF-1 levels relative to IGFBP-3 is that the discrepancies could be due to differences in the specificity of the different assays used for IGFBP-3. All previous studies where IGFBP-3 was related inversely to the risk of cancer after adjustment for IGF-1 (also for cancers of the prostate and breast)[16 24–27] used an ELISA method from Diagnostic Systems Laboratories (DSL, Webster, Texas, USA). The other studies used either an IRMA from Immunotech (Marseille, France)[18 28] or a radioimmunoassay after purification.[29] IGFBP-3 in blood plasma and within tissues undergoes proteolytic cleavage by specific enzymes, including prostate specific antigen and other proteases. It may be that the DSL-ELISA measures more specifically the intact forms of IGFBP-3 whereas the other assays measure the combination of intact and proteolytically cleaved forms. Thus, conceivably, cancer cases could have higher levels of total IGFBP-3 (intact plus proteolytically cleaved forms) which would be reflected by the Immunotech-IRMA method, but also have more proteolytic cleavage of IGFBP-3 (and hence lower levels of intact IGFBP-3) which would be reflected more by the DSL-ELISA method. We recently tested this hypothesis in a study of 102 cases of colorectal cancer and 200 matched controls nested within the New York University Women's Health Study comparing IGFBP-3 measurements obtained by the Immunotech-IRMA and DSL-ELISA. We found that the two types of IGFBP-3 measurements were highly correlated (Spearman's correlations 0.82), and that neither IGFBP-3 assay demonstrated any clear increase in risk with increasing levels of IGF-1 adjusting for IGFBP-3, or any inverse association of risk with IGFBP-3 adjusting for IGF-1. These results thus argue against the hypothesis of different assay specificities, but should be confirmed by further studies.

GH is a principal regulator of plasma and tissue levels of both IGF-1 and IGFBP-3, and increases in pituitary GH secretion during the pubertal growth spurt[30 31] or in pathological conditions such as acromegaly[32 33] are generally accompanied by rises in circulating IGF-1 and IGFBP-3. However, the rise in IGF-1 is generally stronger than that in IGFBP-3 and hence the IGF-1/IGFBP-3 ratio is usually also increased under conditions of elevated GH secretion.[32 34] We therefore speculate that the observations in this and other studies relating elevated circulating IGF-1 levels to an increased risk of colon cancer, either as absolute concentrations or as concentrations relative to IGFBP-3, might reflect a relative increase in pituitary GH secretion.

The inverse (but non-significant) associations of both plasma IGF-1 and IGFBP-3 with the risk of rectal cancer in the present study is puzzling, and does not fit our hypothesis that elevated IGF-1 levels would generally enhance tumour development. This observation is even more striking in view of the positive association of IGF-1 levels with the risk of colon cancer within the same study. The inverse association of IGF-1 with rectal cancer risk could not be explained by confounding by smoking, height, BMI, or IGFBP-3 levels. The total number of rectal cancer cases was small however and further studies are needed to confirm whether this inverse association reflects a true relationship or is due to chance.

It has been suggested that IGF-1 may predict more strongly the risk of advanced disease status at presentation. For example, in a large prospective study of prostate cancer risk in relation to prediagnostic serum IGF-1 levels, the associations of IGF-1 and IGFBP-3 were limited to the presentation of more advanced disease.[35] Our data confirmed the general observation that rectal cancers tend to present at an earlier stage than colon cancers, and this may have masked associations with IGF-1 levels. However, in none of the Dukes' stage categories (A, B, or C) were IGF-1 levels related to cancer risk when cancers of the colon and rectum were combined. We thus conclude that further larger studies are needed to examine the association of IGF-1 with colon cancer and rectal cancer separately, and to estimate separately the relationship between IGF-1 and the risk of advanced or less advanced tumours.

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### Authors' affiliations

**R Palmqvist, R Stenling,** Department of Medical Biosciences, Pathology, Umeå University, SE-901 87 Umeå, Sweden
**G Hallmans,** Department of Public Health and Clinical Medicine, Nutritional Research, Umeå University, SE-901 87 Umeå, Sweden
**S Rinaldi, C Biessy, E Riboli, R Kaaks,** International Agency for Research on Cancer, 150 Cours Albert Thomas, 69372 Lyon Cedex 08, France

## REFERENCES

1 **Werner H**, LeRoith D. The role of the insulin-like growth factor system in human cancer. *Adv Cancer Res* 1996;**68**:183–223.
2 **Giovannucci E**. Insulin and colon cancer. *Cancer Causes Control* 1995;**6**:164–79.
3 **Singh P**, Rubin N. Insulinlike growth factors and binding proteins in colon cancer. *Gastroenterology* 1993;**105**:1218–37.
4 **Freier S**, Weiss O, Eran M, *et al*. Expression of the insulin-like growth factors and their receptors in adenocarcinoma of the colon. *Gut* 1999;**44**:704–8.
5 **Lahm H**, Suardet L, Laurent PL, *et al*. Growth regulation and co-stimulation of human colorectal cancer cell lines by insulin-like growth factor I, II and transforming growth factor alpha. *Br J Cancer* 1992;**65**:341–6.
6 **Lahm H**, Amstad P, Wyniger J, *et al*. Blockade of the insulin-like growth-factor-I receptor inhibits growth of human colorectal cancer cells: evidence of a functional IGF-1I-mediated autocrine loop. *Int J Cancer* 1994;**58**:452–9.
7 **Sell C**, Dumenil G, Deveaud C, *et al*. Effect of a null mutation of the insulin-like growth factor I receptor gene on growth and transformation of mouse embryo fibroblasts. *Mol Cell Biol* 1994;**14**:3604–12.
8 **Cats A**, Dullaart RP, Kleibeuker JH, *et al*. Increased epithelial cell proliferation in the colon of patients with acromegaly. *Cancer Res* 1996;**56**:523–6.
9 **Colao A**, Balzano A, Ferone D, *et al*. Increased prevalence of colonic polyps and altered lymphocyte subset pattern in the colonic lamina propria in acromegaly. *Clin Endocrinol (Oxf)* 1997;**47**:23–8.
10 **Orme SM**, McNally RJ, Cartwright RA, *et al*. Mortality and cancer incidence in acromegaly: a retrospective cohort study. United Kingdom Acromegaly Study Group. *J Clin Endocrinol Metab* 1998;**83**:2730–4.
11 **Jenkins PJ**, Fairclough PD, Richards T, *et al*. Acromegaly, colonic polyps and carcinoma. *Clin Endocrinol (Oxf)* 1997;**47**:17–22.
12 **Jenkins PJ**, Frajese V, Jones AM, *et al*. Insulin-like growth factor I and the development of colorectal neoplasia in acromegaly. *J Clin Endocrinol Metab* 2000;**85**:3218–21.
13 **Jones JI**, Clemmons DR. Insulin-like growth factors and their binding proteins: biological actions. *Endocr Rev* 1995;**16**:3–34.
14 **Thissen JP**, Ketelslegers JM, Underwood LE. Nutritional regulation of the insulin-like growth factors. *Endocr Rev* 1994;**15**:80–101.
15 **Kaaks R**, Lukanova A. Energy balance and cancer: the role of insulin and insulin-like growth factor-I. *Proc Nutr Soc* 2001;**60**:91–106.
16 **Ma J**, Pollak MN, Giovannucci E, *et al*. Prospective study of colorectal cancer risk in men and plasma levels of insulin-like growth factor (IGF)-I and IGF-binding protein-3. *J Natl Cancer Inst* 1999;**91**:620–5.
17 **Giovannucci E**, Pollak MN, Platz EA, *et al*. A prospective study of plasma insulin-like growth factor-1 and binding protein-3 and risk of colorectal neoplasia in women. *Cancer Epidemiol Biomarkers Prev* 2000;**9**:345–9.
18 **Kaaks R**, Toniolo P, Akhmedkhanov A, *et al*. Serum C-peptide, insulin-like growth factor (IGF)-I, IGF-binding proteins, and colorectal cancer risk in women. *J Natl Cancer Inst* 2000;**92**:1592–600.
19 **Ying S**. The PHREG procedure. In: Kelly D, ed. *SAS technical report P-229, SAS/SAT software: changes and enhancements.* Cary (NC): SAS Institute, 1992:433–81.
20 **Manousos O**, Souglakos J, Bosetti C, *et al*. IGF-1 and IGF-1I in relation to colorectal cancer. *Int J Cancer* 1999;**83**:15–7.
21 **Williams AC**, Collard TJ, Perks CM, *et al*. Increased p53-dependent apoptosis by the insulin-like growth factor binding protein IGFBP-3 in human colonic adenoma-derived cells. *Cancer Res* 2000;**60**:22–7.
22 **Gill ZP**, Perks CM, Newcomb PV, *et al*. Insulin-like growth factor-binding protein (IGFBP-3) predisposes breast cancer cells to programmed cell death in a non-IGF-dependent manner. *J Biol Chem* 1997;**272**:25602–7.
23 **Rajah R**, Khare A, Lee PD, *et al*. Insulin-like growth factor-binding protein-3 is partially responsible for high-serum-induced apoptosis in PC-3 prostate cancer cells. *J Endocrinol* 1999;**163**:487–94.
24 **McKeown-Eyssen G**. Epidemiology of colorectal cancer revisited: are serum triglycerides and/or plasma glucose associated with risk? *Cancer Epidemiol Biomarkers Prev* 1994;**3**:687–95.
25 **Chan JM**, Stampfer MJ, Giovannucci E, *et al*. Plasma insulin-like growth factor-I and prostate cancer risk: a prospective study. *Science* 1998;**279**:563–6.
26 **Hankinson SE**, Willett WC, Colditz GA, *et al*. Circulating concentrations of insulin-like growth factor-I and risk of breast cancer. *Lancet* 1998;**351**:1393–6.
27 **Ma J**, Pollak MN, Giovannucci E, *et al*. Prospective study of colorectal cancer risk in men and plasma levels of insulin-like growth factor (IGF)-I and IGF-binding protein-3. *J Natl Cancer Inst* 1999;**91**:620–5.
28 **Stattin P**, Bylund A, Rinaldi S, *et al*. Plasma insulin-like growth factor-I, insulin-like growth factor-binding proteins, and prostate cancer risk: a prospective study. *J Natl Cancer Inst* 2000;**92**:1910–7.
29 **Toniolo P**, Bruning PF, Akhmedkhanov A, *et al*. Serum insulin-like growth factor-I and breast cancer. *Int J Cancer* 2000;**88**:828–32.
30 **Veldhuis JD**, Iranmanesh A. Physiological regulation of the human growth hormone (GH)-insulin-like growth factor type I (IGF-1) axis: predominant impact of age, obesity, gonadal function, and sleep. *Sleep* 1996;**19**:S221–4.
31 **Hesse V**, Jahreis G, Schambach H, *et al*. Insulin-like growth factor I correlations to changes of the hormonal status in puberty and age. *Exp Clin Endocrinol* 1994;**102**:289–98.
32 **Juul A**, Main K, Blum WF, *et al*. The ratio between serum levels of insulin-like growth factor (IGF)-I and the IGF binding proteins (IGFBP-1, 2 and 3) decreases with age in healthy adults and is increased in acromegalic patients. *Clin Endocrinol (Oxf)* 1994;**41**:85–93.
33 **Jasper H**, Pennisi P, Vitale M, *et al*. Evaluation of disease activity by IGF-1 and IGF binding protein-3 (IGFBP3) in acromegaly patients distributed according to a clinical score. *J Endocrinol Invest* 1999;**22**:29–34.
34 **Juul A**, Flyvbjerg A, Frystyk J, *et al*. Serum concentrations of free and total insulin-like growth factor-I, IGF binding proteins -1 and -3 and IGFBP-3 protease activity in boys with normal or precocious puberty. *Clin Endocrinol (Oxf)* 1996;**44**:515–23.
35 **Chan JM**, Stampfer MJ, Giovannucci E, *et al*. Insulin-like growth factor I (IGF-1), IGF-binding protein-3 and prostate cancer risk: epidemiological studies. *Growth Horm IGF Res* 2000;**10**(Suppl A):S32–3.