

Insulin-Like Growth Factor I, IGF-Binding Protein 3, and Lung Cancer Risk in a Prospective Study of Men in China

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Background: Insulin-like growth factor I (IGF-I) stimulates cell proliferation and inhibits apoptosis in the lung and other tissues by interacting with the IGF-I receptor. The major binding protein for IGF-I, insulin-like growth factor-binding protein 3 (IGFBP-3), modulates the effects of IGF-I but also inhibits cell growth and induces apoptosis independent of IGF-I and its receptor. In a prospective study of men in Shanghai, China, we examined the association between serum levels of IGF-I and IGFBP-3 and the subsequent risk of lung cancer. **Methods:** From 1986 to 1989, serum was collected from 18 244 men aged 45–64 years living in Shanghai without a history of cancer. We analyzed IGF-I and IGFBP-3 levels in serum from 230 case patients who developed incident lung cancer during follow-up and from 740 control subjects. **Results:** Among 230 case patients and 659 matched control subjects, increased IGF-I levels were not associated with increased risk of lung cancer. However, for subjects in the highest quartile relative to the lowest quartile of IGFBP-3, the odds ratio (OR) for lung cancer, adjusted for smoking and IGF-I, was 0.50 (95% confidence interval [CI] = 0.25 to 1.02). When the analysis was restricted to ever smokers (184 case patients and 344 matched control subjects), the OR for lung cancer in men in the highest quartile of IGFBP-3 relative to those in the lowest quartile, adjusted for smoking and IGF-I, was 0.41 (95% CI = 0.18 to 0.92). **Conclusions:** In this prospective study of Chinese men, higher serum levels of IGF-I did not increase the risk of lung cancer. However, subjects with higher serum levels of IGFBP-3 were at reduced risk of lung cancer. This finding is consistent with experimental data that indicate that IGFBP-3 can inhibit cellular proliferation and induce apoptosis independent of

IGF-I and the IGF-I receptor. [J Natl Cancer Inst 2002;94:749–54]

Insulin-like growth factor I (IGF-I) is a single-chain polypeptide involved in the regulation of many cellular functions, including proliferation and differentiation (1). It is mitogenic in many tissues and inhibits apoptosis. The effects of IGF-I are mediated primarily through its interaction with the IGF-I receptor. Blocking interaction with this receptor can reverse IGF-I-stimulated proliferation of lung cancer cells (2,3). In the circulation and in the extracellular space, almost all IGF-I is bound to its major binding protein, insulin-like growth factor-binding protein 3 (IGFBP-3). *In vivo* administration of IGF-I has a number of anabolic effects, including increased glucose uptake and increased protein synthesis (1).

IGFBP-3 regulates the biological activities of IGF-I in several ways: it increases the half-life of IGF-I, it enables localization of IGF-I to specific tissues and cell types, and it modulates IGF-I's interaction with the IGF-I receptor (1). Although IGFBP-3 can

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potentiate IGF-I's actions under certain circumstances, it generally inhibits the effects of IGF-I by reducing binding to the IGF-I receptor (1). Like circulating IGF-I, circulating IGFBP-3 is produced mainly in the liver (1). However, both IGF-I (4) and IGFBP-3 (5) are also produced locally in many tissues, including in human bronchial epithelial cells.

IGFBP-3 also acts independent of IGF-I and the IGF-I receptor, interacting with a number of other proteins, including the retinoid X-receptor-alpha (RXR- α), transferrin, and most likely, a receptor for IGFBP-3, which has not yet been identified (6,7). IGFBP-3 can both induce apoptosis (7) and inhibit cell growth (8) independent of IGF-I.

Several epidemiologic studies (9) suggest that higher blood levels of IGF-I and/or lower blood levels of IGFBP-3 are associated with an increased risk of various cancers. Two studies (10,11) specifically investigated lung cancer. Lung cancer risk was independently associated with higher levels of IGF-I and lower levels of IGFBP-3 in a case-control study (11), whereas no such associations were observed in a cohort study (10). Because metabolic changes associated with lung cancer and/or its treatment might alter levels of IGF-I and IGFBP-3, cohort studies offer advantages for evaluating the association of lung cancer risk with IGF-I and IGFBP-3 levels (9). We examined the association between prediagnostic serum levels of IGF-I and IGFBP-3 and the risk of subsequent lung cancer among a cohort of men in Shanghai, China.

METHODS

Between January 1986 and September 1989, all men 45 to 64 years of age living in four small, geographically defined areas of the city of Shanghai, China, were invited to participate in a prospective study of diet and cancer (12,13). At recruitment, each subject was interviewed in person by using a structured questionnaire that included questions about demographics, history of tobacco and alcohol use, current diet (45 food items), and medical history. At the completion of the interview, a 10-mL nonfasting blood sample was collected from each study participant. Blood samples were stored on ice immediately after they were collected, the blood was spun, and the serum was frozen at -20°C within 3–4 hours. A total of 18 244 men were enrolled in the study (80% of eligible subjects). All subjects provided written informed consent, and the institutional review boards of the Shanghai Cancer Institute and the University of Southern California approved the protocol.

Follow-up was conducted by annual recontact with all surviving cohort members and by routine reviews of cancer reports from the Shanghai Cancer Registry and death certificates from the local vital statistics offices. Through March 15, 1997, only 120 subjects had been lost to follow-up, and 259 incident cases of lung cancer had been identified. Of the 259 cases, 178 were histopathologically confirmed and 81 were clinically diagnosed on the basis of radiography or computer-assisted tomography. For each case patient with incident lung cancer, three matched control subjects were selected by matching neighborhood of residence, age at interview (within 2 years), and timing of sample collection (month and year). Ninety-four percent of the control subjects had their blood drawn within 1 month of their matched case patient, and all had blood drawn within 4 months.

Laboratory Analyses

IGF-I was measured in serum by radioimmunoassay using a commercial kit from Nichols Institute Diagnostics (San Juan

Capistrano, CA). IGFBP-3 was measured in serum by immunoradiometric assay using a kit from Diagnostic Systems Laboratory (Webster, TX). For each analyte (i.e., IGF-I and IGFBP-3), all samples were analyzed over an 8-week period by using assay kits from a single lot before the assay kit's expiration date. Serum samples were analyzed in matched sets, with each set containing the case patient's sample and the samples from the matched control subjects. Samples within a given matched set were assayed in the same batch by laboratory personnel who were blinded to the case or control status of the serum sample. Duplicate measurements were made for each sample, and the mean of the duplicate measurements was assigned as the sample value. The mean intra-assay coefficient of variation on blind replicate quality control serum samples was 9.4% for IGF-I and 3.2% for IGFBP-3.

Because some serum from the men had been used for previous assays, serum samples were available for only 235 case patients and 750 control subjects. Insufficient serum was available to measure both IGF-I and IGFBP-3 levels for five case patients and 10 control subjects, leaving 230 case patients and 740 control subjects with results for both analytes.

For 96 of the matched case-control sets, the serum samples had undergone a single rapid thaw for a previous assay and then had been quickly refrozen at -20°C until analysis for IGF-I and IGFBP-3. For the remaining 134 matched case-control sets, samples were stored at -20°C continuously from collection until analysis. In a separate pilot study on serum from six men in the same age range as the study subjects, a single freeze-thaw cycle did not additionally contribute to assay variability. In addition, samples from study subjects that had been through a freeze-thaw cycle gave comparable values to samples that had not been through a freeze-thaw cycle for both IGF-I (mean of 126 ng/mL [95% confidence interval {CI} = 121 to 131] for previously thawed samples and 126 ng/mL [95% CI = 122 to 129] for unthawed samples; $P = .83$) and IGFBP-3 (mean of 1841 ng/mL [95% CI = 1793 to 1889] for previously thawed samples versus 1850 ng/mL for unthawed samples [95% CI = 1810 to 1890]; $P = .78$). These results are consistent with published data showing that levels of IGF-I and IGFBP-3 are not altered by up to five freeze-thaw cycles (14). In addition, time in storage was not a significant predictor of levels of either IGF-I or IGFBP-3.

Statistical Analysis

Odds ratios (ORs) and their 95% CIs were calculated with conditional logistic regression (15), retaining the original matching, using Proc PHREG in SAS version 8 (SAS Institute Inc., Cary, NC). We grouped study subjects into quartiles on the basis of the distribution of analyte values in control subjects. Trend tests for exposure-disease associations were based on logistic regression coefficients for continuous terms for IGF-I and IGFBP-3. P values for comparisons of means were calculated by t tests using SAS Proc TTEST for unmatched comparisons and by generalized linear regression using SAS Proc GLM for matched comparisons. All P values quoted are two-sided.

For the analysis of ORs, we deleted 81 control subjects whose matched case patient did not have enough serum sample remaining to measure both analytes. Thus, the conditional logistical regression analysis included 230 case patients and 659 matched control subjects. Of the 230 matched sets, 203 contained three control subjects per case patient, 23 contained two control sub-

jects per case patient, and four contained one control subject per case patient.

In all analyses, we adjusted for smoking status at the time of blood draw by including terms in the conditional logistic regression model for age at starting to smoke, average number of cigarettes smoked per day, and smoking status (never, past, or current). In this cohort, these terms have been shown to best capture the smoking–lung cancer association that is comparable with that observed in other cohort studies (12). Body mass index did not confound the association between IGF-I or IGFBP-3 levels and lung cancer risk and thus was not included in the final models.

We also examined the associations between lung cancer and IGF-I and IGFBP-3 levels among ever smokers. Among the 230 case patients and 659 matched control subjects available for conditional logistic regression analysis, there were 211 case patients and 374 control subjects who had ever smoked. We deleted the 27 smoking case patients and 30 smoking control subjects whose matched set did not contain both a smoking case patient and at least one smoking control subject. Thus, 184 case patients and 344 matched control subjects remained for conditional logistic regression analyses of ever smokers.

RESULTS

Table 1 shows characteristics of the study population. Case patients and control subjects were evenly matched by age at enrollment. Case patients had been diagnosed with incident lung cancer at a median age of 63 years (interquartile range = 59–67 y). As expected, case patients had a greater smoking history than

control subjects. Body mass index, after adjustment for smoking [which is an important determinant of body mass index (16)], did not differ between case patients and control subjects. This Chinese population is substantially lighter than U.S. adult males (17). Serum IGF-I levels differed little between case patients and control subjects (*P* value from matched analysis with 230 case patients and 659 matched control subjects = .36). The mean IGFBP-3 level was slightly lower among case patients (matched *P* = .04).

Among control subjects, serum IGF-I levels correlated moderately strongly with IGFBP-3 levels (Spearman correlation coefficient, *r* = .46, *P* < .001). Age at study enrollment was weakly inversely correlated with serum levels of both IGF-I (*r* = −.16, *P* < .001) and IGFBP-3 (*r* = −.11, *P* = .003). Body mass index was weakly correlated with IGF-I (*r* = 0.12, *P* = .001) and less correlated with IGFBP-3 (*r* = .03, *P* = .36). No statistically significant correlations with height were observed (*r* = .06, *P* = .09 for IGF-I; *r* = .02, *P* = .56 for IGFBP-3). Similarly, the number of cigarettes smoked per day was not statistically significantly correlated with either IGF-I (*r* = −.05, *P* = .15) or IGFBP-3 (*r* = −.04, *P* = .32).

We examined ORs for lung cancer by quartiles of serum IGF-I and IGFBP-3 (Table 2). Higher levels of IGF-I were not associated with an increased risk of lung cancer. A tendency for the highest quartile of IGF-I to confer a lower risk of lung cancer than the other quartiles was reduced after adjustment for both smoking and IGFBP-3. Relative to subjects in the lowest quartile of serum IGFBP-3, those in the highest quartile were at reduced risk of developing lung cancer (Table 2). Adjustment for smoking and IGF-I did not appreciably alter the point estimate (OR = 0.50, 95% CI = 0.25 to 1.02) from the unadjusted model (OR = 0.52, 95% CI = 0.31 to 0.88).

Because smoking leads to repeated exposure to tumor initiators and because the biologic effects of IGF-I and IGFBP-3 influence cellular proliferation and apoptosis (1), the association between IGF-I and IGFBP-3 might differ between smokers and nonsmokers. When we restricted the analysis to ever smokers, IGF-I levels were again not appreciably related to lung cancer risk (Table 3). Ever smokers with higher levels of IGFBP-3 were at reduced risk of lung cancer (OR for individuals in the highest quartile of IGFBP-3 relative to the lowest quartile, adjusted for smoking and IGF-I, was = 0.41, 95% CI = 0.18 to 0.92).

Among ever smokers, further stratification by the number of cigarettes smoked per day generated unstable ORs. However, when ever smokers were divided into those who smoked less than one pack of cigarettes per day and those who smoked one or more packs per day, the same pattern of association was seen in the two groups (data not shown).

Because preclinical disease might alter levels of IGF-I and IGFBP-3, we repeated the analyses, restricting them to the 183 case patients whose diagnosis of lung cancer occurred at least 2 years after enrollment, along with their 520 matched control subjects. In this reduced data set, we again observed no increase in lung cancer risk in association with higher IGF-I and a reduced risk among individuals in the highest quartile of IGFBP-3 (relative to the lowest quartile) after adjustment for smoking and IGF-I (OR = 0.46, 95% CI = 0.20 to 1.05). Case patients who were diagnosed within 2 years of enrollment did not differ appreciably from those diagnosed later in their mean IGF-I levels, (130 ng/mL [95% CI = 112 to 147] versus 122 ng/mL [95% CI = 116 to 128], respectively; *P* = .41) or their mean IGFBP-3

Table 1. Characteristics of the study population by case–control status

Characteristic	Case patients (n = 230)	Control subjects (n = 740)
Age at enrollment (y)		
Median (IQR)*	59 (55 to 62)	59 (55 to 62)
Never smokers, No. (%)†	19 (8.3)	322 (43.5)
Past smokers, No. (%)	19 (8.3)	58 (7.8)
Current smokers, No. (%)	192 (83.5)	360 (48.7)
Among ever smokers (cigarettes/day)		
Median (IQR)	20 (15 to 20)	15 (10 to 20)
Body Mass Index (kg/m ²)		
Mean (95% CI)‡	21.8 (21.4 to 22.2)	22.0 (21.8 to 22.2)
IGF-I, ng/mL§		
Mean (95% CI)	123 (117 to 129)	127 (124 to 129)
Median (IQR)	117 (92 to 146)	124 (99 to 149)
10th percentile	74	77
90th percentile	180	176
IGFBP-3, ng/mL		
Mean (95% CI)	1793 (1730 to 1856)	1863 (1828 to 1898)
Median (IQR)	1726 (1463 to 2053)	1820 (1538 to 2160)
10th percentile	1191	1267
90th percentile	2472	2477
Histology, No. (%)		
Adenocarcinoma	61 (26.5)	
Squamous and small cell	83 (36.1)	
Other and unknown	86 (37.4)	

*IQR = Interquartile range, which is bounded by the 25th and 75th percentiles.

†Percentages may not add up to 100% because of rounding.

‡Adjusted for smoking (past/current/never and cigarettes smoked per day). CI = confidence interval.

§IGF-I = insulin-like growth factor I.

||IGFBP-3 = insulin-like growth factor-binding protein 3.

Table 2. Odds ratios (ORs) and 95% confidence intervals (CIs) for lung cancer by quartiles of serum IGF-I and IGFBP-3 in a prospective study of men in Shanghai, China*

	Quartiles				<i>P</i> _{trend} †
	1	2	3	4	
IGF-I					
Mean, ng/mL	80	111	135	181	
Range, ng/mL	32–99	101–122	124–149	151–479	
Cases/controls	73/167	54/162	52/171	51/159	
OR, unadjusted (95% CI)	1.0 (referent)	0.76 (0.50 to 1.15)	0.68 (0.45 to 1.04)	0.70 (0.45 to 1.10)	.36
OR, smoking adjusted (95% CI)‡	1.0 (referent)	0.68 (0.41 to 1.13)	0.78 (0.48 to 1.27)	0.73 (0.43 to 1.24)	.80
OR, smoking + IGFBP-3 adjusted (95% CI)	1.0 (referent)	0.73 (0.43 to 1.24)	0.86 (0.51 to 1.45)	0.86 (0.47 to 1.57)	.52
IGFBP-3					
Mean, ng/mL	1279	1676	1984	2506	
Range, ng/mL	201–1538	1540–1820	1822–2160	2162–4127	
Cases/controls	67/160	62/173	60/166	41/160	
OR, unadjusted (95% CI)	1.0 (referent)	0.85 (0.56 to 1.28)	0.81 (0.53 to 1.26)	0.52 (0.31 to 0.88)	.04
OR, smoking adjusted (95% CI)	1.0 (referent)	0.88 (0.54 to 1.41)	0.96 (0.59 to 1.56)	0.56 (0.30 to 1.03)	.14
OR, smoking + IGF-1 adjusted (95% CI)	1.0 (referent)	0.84 (0.51 to 1.38)	0.90 (0.52 to 1.54)	0.50 (0.25 to 1.02)	.12

*IGF-I = insulin-like growth factor, IGFBP-3 = insulin-like growth factor binding protein 3.

†*P* trend test based on coefficient for continuous variable in the conditional logistic regression model.

‡Smoking-adjusted models include terms for never, past, or current smoking and number of cigarettes per day.

Table 3. Odds ratios (ORs) and 95% confidence intervals (CIs) for lung cancer by quartiles of serum IGF-I and IGFBP-3 among smokers from a prospective study of men in Shanghai, China*

	Quartiles				<i>P</i> _{trend} †
	1	2	3	4	
IGF-I					
Cases/controls	59/95	41/88	44/84	40/77	
OR, smoking adjusted (95%)‡	1.0 (referent)	0.66 (0.37 to 1.19)	0.84 (0.49 to 1.46)	0.78 (0.42 to 1.42)	.92
OR, smoking + IGFBP-3 adjusted (95% CI)	1.0 (referent)	0.73 (0.39 to 1.36)	0.95 (0.52 to 1.71)	0.94 (0.46 to 1.91)	.33
IGFBP-3					
Cases/controls	56/88	49/94	46/86	33/76	
OR, smoking adjusted (95% CI)	1.0 (referent)	0.85 (0.50 to 1.44)	0.86 (0.49 to 1.49)	0.50 (0.24 to 1.02)	.19
OR, smoking + IGF-I adjusted (95% CI)	1.0 (referent)	0.79 (0.45 to 1.37)	0.75 (0.40 to 1.38)	0.41 (0.18 to 0.92)	.10

*IGF-I = insulin-like growth factor 1; IGFBP-3 = insulin-like growth factor-binding protein 3.

†*P* for trend based on coefficient for continuous variable in the conditional logistic regression model.

‡Smoking-adjusted models include terms for past or current smoking, number of cigarettes per day, and age at starting to smoke.

levels (1754 ng/mL [95% CI = 1610 to 1897] versus 1802 ng/mL [95% CI = 1732 to 1872], respectively; *P* = .54). When matched sets were divided into three categories of time from enrollment to diagnosis (within 4 years, 4–8 years, and >8 years), the same pattern of association was maintained in each category. That is, there was no increased lung cancer risk for individuals with higher levels of IGF-I compared with individuals with the lowest levels of IGF-I, and there was a reduced risk for individuals in the highest category (quartile) of serum IGFBP-3, even though the ORs were unstable within categories (data not shown).

We used histologic information to classify lung cancer cases into three categories—adenocarcinoma, squamous and small cell carcinoma, and other cell types plus those diagnosed on radiologic or clinical grounds (Table 1). Although the small numbers within histologic categories gave unstable ORs, the same pattern of associations persisted in each group: IGF-I was not associated with an increased risk of lung cancer, and subjects with the highest levels of IGFBP-3 were at reduced risk (data not shown).

DISCUSSION

In this prospective study of men in Shanghai, higher serum IGF-I levels were not associated with an increased risk of lung cancer. However, subjects with higher levels of IGFBP-3 were at reduced risk of lung cancer.

Few published studies have addressed lung cancer in relation to IGF-I and IGFBP-3. In a case-control study of 183 case subjects in the United States, higher levels of plasma IGF-I were associated with an increased risk of lung cancer (11,18). However, similar to our findings, subjects with the highest levels of IGFBP-3 were at the lowest risk of lung cancer (OR for highest quartile relative to lowest quartile = 0.48, 95% CI = 0.25 to 0.92) (11). This association between IGFBP-3 and lung cancer risk held for all three histologic cell types and was independent of IGF-I (18). In contrast, in prospective data based on 93 case patients from the New York University (NYU) Women's Health Study, neither serum IGF-I (OR for highest quartile relative to the lowest quartile = 0.54, 95% CI = 0.14 to 2.07) or IGFBP-3

(OR = 0.90, 95% CI = 0.28 to 2.85) showed a statistically significant association with lung cancer (10).

The association between IGF-I and cancer risk may best be evaluated in prospective data because of possible influences of disease and/or its treatment on IGF-I levels (9). Several prospective studies have been carried out on the association between IGF-I and IGFBP-3 and risk of cancers other than lung cancer, although the results are not completely consistent. Plasma IGF-I, without adjustment for IGFBP-3, was statistically significantly positively associated with prostate cancer (19) and breast cancer in younger women (20,21). In some prospective studies, inverse associations of cancer risk with IGFBP-3 have been stronger than the positive associations with IGF-I. For example, in U.S. cohorts of women (22) and men (23), the inverse association between IGFBP-3 and colorectal cancer risk was more marked than the positive association with IGF-I. Furthermore, the statistically significant positive association of colorectal cancer risk with IGF-I was seen only after IGFBP-3 adjustment. However, in two other studies (24,25), higher levels of IGFBP-3 were associated with a statistically significantly increased risk of colorectal cancer, but IGF-I levels showed no such association. IGFBP-3 was also positively related to breast cancer risk among women under age 50 years in the NYU Women's Health Study (20).

The number of subjects prospectively studied with respect to IGF-I, IGFBP-3, and cancer risk remains relatively small, and inconsistencies between studies may arise from chance. The use of different analytic procedures does not appear to explain the heterogeneity in results. Although differences in the distributions of levels of circulating IGF-I and IGFBP-3 between populations might contribute to these inconsistencies, it is impossible to evaluate this hypothesis because absolute levels of these analytes cannot be compared across studies. The commercial assay kits that are used tend to give different absolute values in the same populations over time. Thus, different groups of samples from control subjects in the same cohort, measured over time for comparison with case-patients with different cancers, do not give identical levels of IGF-I and/or IGFBP-3, despite similar age distributions. This variability has been seen in all of the cohorts in which the association between IGF-I, IGFBP-3, and risk of various cancers has been studied prospectively. These studies include the Physicians' Health Study (19,23), the Nurses' Health Study (21,26,27), the NYU Women's Health Study (10,20,25), and this Shanghai cohort (24). Groups can only be compared if their samples are measured together within the same analytic batches. However, because in all of these studies careful attention has been paid to analyzing case samples and their matched control samples in the same analytic batch, and they presumably used reagents from a single lot as we did, internal comparisons should be valid.

Potential limitations of this study, as well as of other studies of IGF-I and IGFBP-3 and cancer risk, include the use of a single measurement to classify individuals. This and other sources of measurement error such as laboratory variability will generally attenuate associations. However, in adults of the same age range as our study subjects, blood samples drawn 0.75–4.75 years apart produced highly correlated values for both IGF-I ($r = .87$) and IGFBP-3 ($r = .73$) (25). With respect to laboratory variability, we measured IGF-I and IGFBP-3 levels in serum, not plasma, which was measured in some other studies. However, Yu et al. (11) found that serum and plasma gave similar

values for both IGF-I and IGFBP-3 (14). Our samples had been stored for more than 10 years at -20°C , and thus some degradation may have occurred, contributing to measurement error. However, the moderately strong correlation we observed between IGF-I and IGFBP-3 is comparable with that reported in other studies (11,19,22,25). In addition, the degree of laboratory variability (as determined by intra-assay coefficient of variability) in our study was comparable with that in other published reports. Thus, we do not believe that measurement error was a greater limitation in this study than in most others.

The association between IGF-I and cancer risk might be expected to vary by cancer site. In prospective studies, positive associations have been reported between IGF-I and cancers of the breast, colon, and prostate that have been related to factors such as body size, energy balance (28), and physical activity (29), either in adulthood or early life (30). In adults, IGF-I is not generally related to height and is only weakly related to adult body mass index in a nonlinear fashion (31). However, before puberty, IGF-I closely reflects both height and body mass index (32). If associations between risk of breast, prostate, and colon cancer and IGF-I reflect some aspect of early growth, nutrition, or energy balance (33), weaker associations would be predicted for lung cancer, for which smoking is the major risk factor and evidence for an important independent role of energy balance or body size is much less compelling. The association between IGF-I and lung cancer risk might differ between our cohort of Chinese men in Shanghai and the U.S. population studied by Yu et al. (11) because of disparities in patterns of early or later growth or nutrition that may influence circulating levels of IGF-I.

The finding of decreased risk of lung cancer with higher levels of IGFBP-3, without a positive association with IGF-I, is biologically plausible. A growing literature has identified biologic effects of IGFBP-3 that are independent of IGF-I and/or the IGF-I receptor. These include induction of apoptosis (7), inhibition of cell growth (8), mediation of $1\alpha,25$ -dihydroxyvitamin D_3 -induced growth inhibition *in vitro* (34), and modulation of growth arrest and apoptosis after oxidant exposure in a human lung epithelial cell line (35). IGFBP-3 binds to a number of proteins other than IGF-I, including RXR- α and transferrin, and efforts are in progress to characterize the IGFBP-3 receptor (7). IGFBP-3 is also known to be transported to the nucleus (36). Although the role of nuclear IGFBP-3 is unknown, it might modulate the activity of nuclear transcription factors, have a specific signal transduction role in the nucleus, and/or regulate gene expression (37). Future work will likely elucidate additional actions of IGFBP-3 relevant to carcinogenesis.

In summary, in this prospective cohort of men in Shanghai, higher serum levels of IGF-I were not associated with an increased risk of lung cancer. However, higher serum levels of IGFBP-3, which has antiproliferative effects that are independent of IGF-I and the IGF-I receptor, was inversely associated with lung cancer risk.

REFERENCES

- (1) Jones JJ, Clemmons DR. Insulin-like growth factors and their binding proteins. Biological actions. *Endocr Rev* 1995;16:3–34.
- (2) Favoni RE, de Cupis A, Ravera F, Cantoni C, Pirani P, Ardizzoni A, et al. Expression and function of the insulin-like growth factor I system in human non-small-cell lung cancer and normal lung cell lines. *Int J Cancer* 1994; 56:858–66.

- (3) Ankrapp DP, Bevan DR. Insulin-like growth factor-I and human lung fibroblast-derived insulin-like growth factor-I stimulate the proliferation of human lung carcinoma cells *in vitro*. *Cancer Res* 1993;53:3399-404.
- (4) Zhang S, Smartt H, Holgate ST, Roche WR. Growth factors secreted by bronchial epithelial cells control myofibroblast proliferation: an *in vitro* co-culture model of airway remodeling in asthma. *Lab Invest* 1999;79:395-405.
- (5) Han GR, Dohi DF, Lee HY, Rajah R, Walsh GL, Hong WK, et al. All-trans-retinoic acid increases transforming growth factor-beta2 and insulin-like growth factor binding protein-3 expression through a retinoic acid receptor-alpha-dependent signaling pathway. *J Biol Chem* 1997;272:13711-6.
- (6) Weinzimer SA, Gibson TB, Collett-Solberg PF, Khare A, Liu B, Cohen P. Transferrin is an insulin-like growth factor-binding protein-3 binding protein. *J Clin Endocrinol Metab* 2001;86:1806-13.
- (7) Liu B, Lee HY, Weinzimer SA, Powell DR, Clifford JL, Kurie JM, et al. Direct functional interactions between insulin-like growth factor-binding protein-3 and retinoid X receptor-alpha regulate transcriptional signaling and apoptosis. *J Biol Chem* 2000;275:33607-13.
- (8) Oh Y, Muller HL, Lamson G, Rosenfeld RG. Insulin-like growth factor (IGF)-independent action of IGF-binding protein-3 in Hs578T human breast cancer cells. Cell surface binding and growth inhibition. *J Biol Chem* 1993;268:14964-71.
- (9) Yu H, Rohan T. Role of the insulin-like growth factor family in cancer development and progression. *J Natl Cancer Inst* 2000;92:1472-89.
- (10) Lukanova A, Toniolo P, Akhmedkhanov A, Biessy C, Haley NJ, Shore RE, et al. A prospective study of insulin-like growth factor-I, IGF-binding proteins-1, -2 and -3 and lung cancer risk in women. *Int J Cancer* 2001;92:888-92.
- (11) Yu H, Spitz MR, Mistry J, Gu J, Hong WK, Wu X. Plasma levels of insulin-like growth factor-I and lung cancer risk: a case-control analysis. *J Natl Cancer Inst* 1999;91:151-6.
- (12) Yuan JM, Ross RK, Wang XL, Gao YT, Henderson BE, Yu MC. Morbidity and mortality in relation to cigarette smoking in Shanghai, China. A prospective male cohort study. *JAMA* 1996;275:1646-50.
- (13) Ross RK, Yuan JM, Yu MC, Wogan GN, Qian GS, Tu JT, et al. Urinary aflatoxin biomarkers and risk of hepatocellular carcinoma. *Lancet* 1992;339:943-6.
- (14) Yu H, Mistry J, Nicar MJ, Khosravi MJ, Diamandis A, van Doorn J, et al. Insulin-like growth factors (IGF-I, free IGF-I and IGF-II) and insulin-like growth factor binding proteins (IGFBP-2, IGFBP-3, IGFBP-6, and ALS) in blood circulation. *J Clin Lab Anal* 1999;13:166-72.
- (15) Breslow NE, Day NE. Statistical methods in cancer research. Volume I—the analysis of case-control studies. IARC Sci Publ 1980;32:5-338.
- (16) Wannamethee S, Shaper A, Walker M. Weight change, body weight and mortality: the impact of smoking and ill health. *Int J Epidemiol* 2001;30:777-86.
- (17) Eberhardt MS, Ingram DD, Makuc DM, et al. Urban and Rural Health Chartbook. Health, United States, 2001. Hyattsville, MD: National Center for Health Statistics, 2001.
- (18) Wu X, Yu H, Amos CI, Hong WK, Spitz MR. Joint effect of insulin-like growth factors and mutagen sensitivity in lung cancer risk. *J Natl Cancer Inst* 2000;92:737-43.
- (19) Chan JM, Stampfer MJ, Giovannucci E, Gann PH, Ma J, Wilkinson P, et al. Plasma insulin-like growth factor-I and prostate cancer risk: a prospective study. *Science* 1998;279:563-6.
- (20) Toniolo P, Bruning PF, Akhmedkhanov A, Bonfrer JM, Koenig KL, Lukanova A, et al. Serum insulin-like growth factor-I and breast cancer. *Int J Cancer* 2000;88:828-32.
- (21) Hankinson SE, Willett WC, Colditz GA, Hunter DJ, Michaud DS, Deroo B, et al. Circulating concentrations of insulin-like growth factor-I and risk of breast cancer. *Lancet* 1998;351:1393-6.
- (22) Giovannucci E, Pollak MN, Platz EA, Willett WC, Stampfer MJ, Majeed N, et al. A prospective study of plasma insulin-like growth factor-I and binding protein-3 and risk of colorectal neoplasia in women. *Cancer Epidemiol Biomarkers Prev* 2000;9:345-9.
- (23) Ma J, Pollak MN, Giovannucci E, Chan JM, Tao Y, Hennekens CH, 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.
- (24) Probst-Hensch NM, Yuan JM, Stanczyk FZ, Gao YT, Ross RK, Yu MC. IGF-1, IGF-2 and IGFBP-3 in prediagnostic serum: association with colorectal cancer in a cohort of Chinese men in Shanghai. *Br J Cancer* 2001;85:1695-99.
- (25) Kaaks R, Toniolo P, Akhmedkhanov A, Lukanova A, Biessy C, Dechaud H, 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.
- (26) Giovannucci E, Pollak M, Platz EA, Willett WC, Stampfer MJ, Majeed N, et al. Insulin-like growth factor I (IGF-I), IGF-binding protein-3 and the risk of colorectal adenoma and cancer in the Nurses' Health Study. *Growth Horm IGF Res* 2000;10 Suppl A:S30-1.
- (27) Byrne C, Colditz GA, Willett WC, Speizer FE, Pollak M, Hankinson SE. Plasma insulin-like growth factor (IGF) I, IGF-binding protein 3, and mammographic density. *Cancer Res* 2000;60:3744-8.
- (28) Visscher TL, Seidell JC. The public health impact of obesity. *Annu Rev Public Health* 2001;22:355-75.
- (29) Friedenreich CM. Physical activity and cancer prevention: from observational to intervention research. *Cancer Epidemiol Biomarkers Prev* 2001;10:287-301.
- (30) Frankel S, Gunnell DJ, Peters TJ, Maynard M, Davey Smith G. Childhood energy intake and adult mortality from cancer: the Boyd Orr Cohort Study. *BMJ* 1998;316:499-504.
- (31) Copeland KC, Colletti RB, Devlin JT, McAuliffe TL. The relationship between insulin-like growth factor-I, adiposity, and aging. *Metabolism* 1990;39:584-7.
- (32) Juul A, Bang P, Hertel NT, Main K, Dalgaard P, Jorgensen K, et al. Serum insulin-like growth factor-I in 1030 healthy children, adolescents, and adults: relation to age, sex, stage of puberty, testicular size, and body mass index. *J Clin Endocrinol Metab* 1994;78:744-52.
- (33) Holly JM, Gunnell DJ, Davey Smith G. Growth hormone, IGF-I and cancer. Less intervention to avoid cancer? More intervention to prevent cancer? *J Endocrinol* 1999;162:321-30.
- (34) Boyle BJ, Zhao XY, Cohen P, Feldman D. Insulin-like growth factor binding protein-3 mediates 1 alpha,25-dihydroxyvitamin d(3) growth inhibition in the LNCaP prostate cancer cell line through p21/WAF1. *J Urol* 2001;165:1319-24.
- (35) Besnard V, Corroyer S, Trugnan G, Chadelat K, Nabeyrat E, Cazals V, et al. Distinct patterns of insulin-like growth factor binding protein (IGFBP)-2 and IGFBP-3 expression in oxidant exposed lung epithelial cells. *Biochim Biophys Acta* 2001;1538:47-58.
- (36) Schedlich LJ, Young TF, Firth SM, Baxter RC. Insulin-like growth factor-binding protein (IGFBP)-3 and IGFBP-5 share a common nuclear transport pathway in T47D human breast carcinoma cells. *J Biol Chem* 1998;273:18347-52.
- (37) Schedlich LJ, Le Page SL, Firth SM, Briggs LJ, Jans DA, Baxter RC. Nuclear import of insulin-like growth factor-binding protein-3 and -5 is mediated by the importin beta subunit. *J Biol Chem* 2000;275:23462-70.

NOTES

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