Dietary Correlates of Plasma Insulin-like Growth Factor I and Insulin-like Growth Factor Binding Protein 3 Concentrations[1]

Michelle D. Holmes,[2] Michael N. Pollak, Walter C. Willett, and Susan E. Hankinson

Channing Laboratory, Department of Medicine, Harvard Medical School and Brigham and Women's Hospital, Boston, Massachusetts 02115 [M. D. H., W. C. W., S. E. H.]; Cancer Prevention Research Unit, Department of Oncology, Jewish General Hospital and McGill University, Montreal, Quebec, H3T1E2 Canada [M. N. P.]; Department of Epidemiology, Harvard School of Public Health, Boston, Massachusetts 02115 [W. C. W. and S. E. H.]; and Department of Nutrition, Harvard School of Public Health, Boston, Massachusetts 02115 [W. C. W.]

Abstract

Plasma levels of insulin-like growth factor I (IGF-I) have been associated with risk of several cancers. Although protein-calorie malnutrition is known to decrease IGF-I levels, few published studies have related diet to IGF-I levels in well-nourished humans. We examined the cross-sectional association of plasma IGF-I and IGF-binding protein 3 (IGFBP-3) levels with intakes of alcohol, energy, macronutrients, micronutrients, and specific foods in 1037 healthy women. Adjusted mean hormone levels across categories of dietary variables were calculated by linear regression. Results were adjusted for non-dietary factors found to be associated with IGF levels. Total energy intake was positively associated with IGF-I levels when adjusted for covariates. Adjusted mean levels of IGF-I (ng/ml) across increasing quintiles of energy intake were 181, 185, 191, 199, and 195 (P for the linear trend = 0.006). In other multivariate analyses, energy-adjusted fat and carbohydrate intake had no association with IGF-I levels. The most consistent finding was a positive association between protein intake with circulating IGF-I concentration (174, 188, 201, 192, and 196 ng/ml across quintiles of protein intake; $P = 0.002$), which was largely attributable to milk intake. Adjusted mean levels of IGF-I (ng/ml) across increasing quartiles of milk intake were 183, 189, 188, and 200 ($P = 0.01$). Higher fat intake, in particular saturated fat, was associated with lower levels of IGFBP-3. We conclude that higher energy, protein, and milk intakes were associated with higher levels of IGF-I. These associations raise the possibility that diet could affect cancer risk through influencing IGF-I level.

Introduction

IGFs[3] are important in the growth and function of many organs (1). Circulating IGF-I concentration is a complex trait influenced by both genetic and nongenetic factors, but these have not yet been characterized in detail (2). There is considerable interindividual variation in plasma levels of the peptide hormone IGF-I, and differences in circulating IGF-I levels have been associated with risk of diseases. There is some evidence that higher plasma levels of IGF-I are associated with lower risk of osteoporosis (3) and heart disease (4). Several studies provide evidence that higher circulating levels of IGF-I and/or lower levels of the principal IGF binding protein, IGFBP-3, are associated with increased risk of prostate cancer (5–7), colon cancer (8–11), lung cancer (12), and premenopausal breast cancer (13–15). Several of these studies provide prospective evidence for cancer risk (5, 7–9, 13, 15). The biological plausibility of the IGF-cancer link has been reviewed recently (16).

It is well established that protein-calorie malnutrition lowers IGF levels (17); however, little is known about how diet affects IGF levels in well-nourished humans. One study reported that IGF-I levels were positively associated with intake of red meat and total fat and inversely associated with intake of carbohydrates (18). In other studies, positive associations have been seen between IGF-I levels and alcohol (19) and zinc (20).

Knowledge of dietary factors associated with IGF levels may elucidate disease mechanisms and offer methods of disease prevention. In the present study, we examined the cross-sectional association of IGF-I and IGFBP-3 levels with intakes of various dietary factors in 1037 healthy women enrolled in the NHS. In another report, we address the association of IGFs with age, BMI, smoking, physical activity, and reproductive and menopausal factors (21).

Materials and Methods

The methods for the collection of blood samples and laboratory assays are described in detail elsewhere (13, 21). Briefly, the study participants were women in the NHS from whom blood samples were collected in 1989 or 1990. Participants were 43–69 years of age at the time. Samples used in this analysis were frozen and stored and later assayed in two batches in 1996 and 1998. This study was approved by the Institutional Review

Received 6/1/01; revised 5/7/02; accepted 5/19/02.
The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked *advertisement* in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
[1] Supported by NIH Grants CA87969 and CA49449. S. E. H. was partially supported by Career Development Award DAMD17-96-1-6021 from the United States Army Medical Research and Materiel Command. M. N. P. was supported by the National Cancer Institute of Canada and the Canadian Breast Cancer Research Initiative and by a grant from the Streams of Excellence program of the Canadian Breast Cancer Research Initiative. M. D. H. was partially funded by the National Cancer Institute Specialized Program of Research Excellence in Breast Cancer at Brigham and Women's Hospital.
[2] To whom requests for reprints should be addressed, at Channing Laboratory, Department of Medicine, Harvard Medical School and Brigham and Women's Hospital, 181 Longwood Avenue, Boston, MA 02115. Phone: (617) 525-2747; Fax: (617) 525-2008; E-mail: michelle.holmes@channing.harvard.edu.

[3] The abbreviations used are: IGF, insulin-like growth factor; IGFBP-3, IGF binding protein 3; NHS, Nurses' Health Study; BMI, body mass index; FFQ, food frequency questionnaire; HRT, hormone replacement therapy; GH, growth hormone; rBGH, recombinant bovine GH.

Board of the Brigham and Women's Hospital. IGF-I and IGFBP-3 were assayed by ELISA with reagents from Diagnostic Systems Laboratory (Webster, TX). Details of the assays for IGF-I and IGFBP-3 levels have been reported previously (13, 21).

Women included in this analysis were controls in a nested case-control study of plasma hormone levels and breast cancer risk (13). In addition, 49 women who had reported very low fat intake (<25 and <19% of energy on the 1986 and 1990 FFQs, respectively) were added to increase the range of fat intake. These cutpoints were chosen to maximize the number of women with low fat intakes on both questionnaires, and all women meeting these criteria were included in this analysis. Because fat intake decreased in the entire cohort over time, we were able to use a more stringent criteria for the later (1990) FFQ. These samples were interspersed with the control and were assayed at the same time. Women defined as postmenopausal had no menses for at least 12 months before blood sampling. To increase the number of premenopausal women, additional women were chosen according to the following criteria: 50 years of age or less at time of phlebotomy, premenopausal at time of phlebotomy (still having cycles and not taking HRT), not previously in any case-control data set, had sufficient plasma available, and had answered the 1990 diet questionnaire. Of women fitting these criteria, 400 were chosen randomly, and 88 additional premenopausal women with specific characteristics were oversampled: 25 women with a family history of breast cancer, 25 women reporting ≥15 g of alcohol daily in 1990, and 38 women with a reported fat intake <25% of calories in 1986 and ≤20% in 1990, and ≤2 drinks of alcohol/day and total daily energy intake between 1000 and 3000 kilocalories and BMI between 18 and 30 in both 1986 and 1990. Samples from these women were analyzed in the second batch. Participants had no previously diagnosed cancer (except nonmelanoma skin cancer) and could not have implausible scores for total energy intake (<500 kcal or >3500 kcal/day).

In each case, the factor that was oversampled is either hypothesized or known to affect cancer risk. If IGF-I or IGFBP-3 levels are correlated with these factors, then IGF-I or IGFBP-3 might be the mechanism by which these factors affect cancer risk. However, the distribution of these factors in the NHS population is such that a random sample would not provide enough women in the extremes of the distribution to test these associations, and thus we oversampled women in these extreme categories.

The FFQ used has been described in detail, and its validity and reproducibility have been documented elsewhere (22). In this analysis, nutrient intakes from the 1986 and the 1990 FFQs were averaged for each woman. In a previous analysis of dietary intake and steroid hormone levels, we found that associations were stronger for the average of the two dietary assessments than with the dietary assessment closest to blood collection (23). This is presumably because averaging two measurements of dietary intake reduces random error. Alcohol intake was categorized as 0, <5, 5–14.9, and at least 15 g/day. Nutrient and food intakes were energy-adjusted using the residual method (24). Nutrient intakes were categorized into quintiles. Some nutrients, particularly vitamins, are frequently found in nutritional supplements. We report results for the total nutrient, which includes supplement intake, and for the nutrient intake from food only.

Because the FFQ allows only nine choices of intake for foods, the distribution of food intakes is more discontinuous than that of nutrients because nutrients can come from many foods. Therefore, foods were divided into approximate quartiles of intake. We calculated a the glycemic load of a food by multiplying its carbohydrate content by its glycemic index value (25–27); we then multiplied this by the frequency of consumption and summed overall food items to produce the dietary glycemic load. Dietary glycemic load thus represents the quality and quantity of carbohydrates and their interaction, with a higher glycemic index having a greater impact at higher carbohydrate intakes. Each unit of dietary glycemic load represents the equivalent of 1 g of carbohydrate from white bread.

Statistical analyses were done with SAS software (SAS Institute, Cary NC). We excluded 3 women whose hormone values were greater than the absolute value of the 75th percentile plus three times the interquartile range. Adjusted least-square mean hormone levels across categories of factors were calculated by linear regression (28). *P*s are reported for the linear trend across categories. Multivariate models were adjusted for laboratory batch and those factors previously found to be associated with IGF levels: age, menopausal status, type and duration of hormone replacement use, smoking status, physical activity, BMI, parity, and history of ever having breastfed (21).

Results

There were 1037 women included in the study. In 1990, participants in the study had a median (10th–90th percentile) age of 50.5 (45–66) years and a median BMI of 24.1 (21–34) kg/m^2. Median energy intake was 1745 (1205–2398) kcal/day. The distributions of hormone levels and other covariates are described elsewhere (21).

We calculated the age- and energy-adjusted associations of IGF-I and IGFBP-3 with intake of alcohol, energy, macronutrients, vitamins A, C, D, and E, calcium, zinc, caffeine, and seven food groups. The results of multivariate analyses were similar, and only the multivariate results are shown here.

Table 1 shows the multivariate association of alcohol, energy, and nutrients with IGF-I and IGFBP-3. The highest category of alcohol intake was associated with a statistically significant higher IGFBP-3 level compared with no alcohol intake (4042 ng/ml *versus* 3910 ng/ml; $P = 0.02$ for comparison between extreme categories). Higher total energy intake (as measured in kilocalories/day) was associated with higher IGF-I (quintile 5 = 195 ng/ml *versus* 181 ng/ml for quintile 1; *P* for the linear trend = 0.006) and higher IGFBP-3 (quintile 5 = 4092 ng/ml *versus* quintile 1 = 3935 ng/ml; $P = 0.02$).

Higher fat intake was associated with lower IGFBP-3; this association appeared to be primarily attributable to saturated fat. There was little other relation between specific types of fat (animal, vegetable, polyunsaturated, monounsaturated, saturated, *trans*-unsaturated and ω-3 fats from marine sources) and IGF levels. Dietary carbohydrate, glycemic load, and fiber intake were also not associated with IGF-I or IGFBP-3 levels. Higher intake of protein was associated with higher levels of IGF-I. This association was limited to animal protein and did not apply to vegetable protein, possibly because the amount and range of vegetable protein intake was much lower.

We also examined the association of vitamins and other micronutrients with IGF-I and IGFBP-3. Several of these nutrients (vitamin D, calcium, and vitamin A) had a positive association with IGF-I levels only when examining the nutrient intake that came from food sources and not when considering total nutrient intake, which included intake from supplements. For vitamin A, the strongest association was with retinol (preformed vitamin A) from food sources only. Lycopene intake was positively associated with IGFBP-3.

Vitamin E, whether from food sources only or including

Table 1 Multivariate adjusted mean levels of IGF-I, and IGFBP-3, by category of alcohol, energy, and nutrients, and the *P* for the linear trend across categories

All dietary factors were adjusted for: energy intake (quintiles), age in years (<45, 45–49, 50–54, 55–59, 60–64, ≥65), laboratory batch, menopausal status, type, and duration of hormone replacement (premenopausal, uncertain menopausal status, postmenopausal non-hormone user, postmenopausal and oral estrogen user, postmenopausal and oral estrogen plus progesterone use for <5 years, postmenopausal and oral estrogen plus progesterone use for ≥5 years, postmenopausal and other hormone user, missing menopausal status and/or hormone use), smoking status (never, past, current), physical activity in MET-hours/week (<4, 4–11.9, 12–24.9, 25+), parity and history of ever lactating (nulliparous, 1 birth and no lactation, 1 birth and lactation, 2 births and no lactation, 2 births and lactation, 3 births and no lactation, 3 births and lactation, 4+ births and no lactation, 4+ births and lactation, missing parity and/or lactation), and BMI in kg/m^2 (<21, 21–22.9, 23–24.9, 25–28.9, ≥29). Total Vitamin A, total retinol, total vitamin E, total vitamins, and total zinc refer to intake from both supplements and food.

Dietary variable	Category[a] and range of intake	IGF-I (ng/ml)	IGFBP-3 (ng/ml)
Energy (kilocalories/day)			
	Q1 <1383	181	3935
	Q2 1383–1624	185	3944
	Q3 1625–1871	191	3992
	Q4 1872–2154	199	4078
	Q5 ≥2155	195	4092
		$P = 0.006$	$P = 0.02$
Alcohol (g/day)	None	186	3910
	<5	192	4004
	5–14.9	204	4127
	≥15	171	4042
		$P = 0.65$	$P = 0.02$
Total fat (g/day)	Q1 ≤47.8	200	4191
	Q2 47.9–54.3	188	4000
	Q3 54.4–58.8	193	3995
	Q4 58.9–63.6	182	3923
	Q5 >63.6	187	3932
		$P = 0.08$	$P = 0.006$
Animal fat (g/day)	Q1 ≤23.9	187	4080
	Q2 24–28.4	192	4112
	Q3 28.5–32.3	187	3941
	Q4 32.4–36.7	194	3987
	Q5 >36.7	191	3920
		$P = 0.52$	$P = 0.02$
Vegetable fat (g/day)	Q1 ≤19.1	197	3968
	Q2 19.2–23.1	192	4082
	Q3 23.2–26.4	194	4027
	Q4 26.5–30.8	185	3989
	Q5 ≥30.9	184	3977
		$P = 0.04$	$P = 0.63$
Saturated fat (g/day)	Q1 <16.0	198	4158
	Q2 16.0–18.5	187	4057
	Q3 18.6–20.5	189	3951
	Q4 20.6–22.6	194	3982
	Q5 >22.6	184	3892
		$P = 0.28$	$P = 0.005$
Monounsaturated fat (g/day)	Q1 <17.4	198	4127
	Q2 17.4–20.1	194	4045
	Q3 20.2–22.1	181	3941
	Q4 22.2–24.1	188	3996
	Q5 ≥24.2	190	3936
		$P = 0.21$	$P = 0.04$
Polyunsaturated fat (g/day)	Q1 <8.5	198	4001
	Q2 8.5–9.9	190	3982
	Q3 10.0–11.0	184	4032
	Q4 11.1–12.5	193	4044
	Q5 >12.5	186	3982
		$P = 0.22$	$P = 0.92$
trans-Unsaturated fat (g/day)	Q1 <1.8	195	4065
	Q2 1.8–2.2	185	4013
	Q3 2.3–2.6	195	4026
	Q4 2.7–3.2	189	3949
	Q5 >3.2	188	3989
		$P = 0.58$	$P = 0.31$
ω-3 fats from fish (g/day)	Q1 <0.11	178	3922
	Q2 0.11–0.17	197	4011
	Q3 0.18–0.23	190	3955
	Q4 0.24–0.33	190	4040
	Q5 ≥0.34	195	4116
		$P = 0.07$	$P = 0.03$

Table 1 Continued

Dietary variable	Category[a] and range of intake	IGF-I (ng/ml)	IGFBP-3 (ng/ml)
Total protein (g/day)	Q1 <70	174	3913
	Q2 70–72.2	188	4061
	Q3 72.3–77.2	201	4096
	Q4 77.3–83	192	4072
	Q5 >83	196	3900
		$P = 0.002$	$P = 0.93$
Animal protein (g/day)	Q1 <45	173	3895
	Q2 45–51	188	4101
	Q3 52–56	200	4075
	Q4 56–63	198	4097
	Q5 >63	192	3874
		$P = 0.002$	$P = 0.82$
Vegetable protein (g/day)	Q1 ≤17.5	189	3990
	Q2 17.6–19.5	186	3950
	Q3 19.6–21.2	194	3982
	Q4 21.3–23.6	190	4044
	Q5 >23.6	193	4077
		$P = 0.49$	$P = 0.17$
Carbohydrate (g/day)	Q1 ≤176	181	3933
	Q2 177–190	195	3996
	Q3 191–203	192	4056
	Q4 204–221	193	3997
	Q5 >221	190	4059
		$P = 0.29$	$P = 0.21$
Fiber (g/day)	Q1 ≤13.8	183	3997
	Q2 13.9–16.0	185	3969
	Q3 16.1–18.4	195	3988
	Q4 18.5–21.5	196	4053
	Q5 >21.5	192	4034
		$P = 0.08$	$P = 0.43$
Glycemic load (units/day)	Q1 <128	178	3914
	Q2 128–141	197	4018
	Q3 142–153	191	4069
	Q4 154–168	194	4008
	Q5 >168	191	4033
		$P = 0.10$	$P = 0.24$
Vitamin D (IU/day)	Q1 <156	189	3969
	Q2 156–240	182	3996
	Q3 241–345	193	3969
	Q4 346–494	193	3927
	Q5 >494	195	4180
		$P = 0.12$	$P = 0.07$
Vitamin D, food sources only (IU/day)	Q1 ≤116	185	3952
	Q2 117–157	184	3972
	Q3 158–202	184	4007
	Q4 203–266	200	4070
	Q5 >266	198	4040
		$P = 0.006$	$P = 0.15$
Calcium (mg/day)	Q1 <672	190	4006
	Q2 672–873	187	4029
	Q3 874–1072	190	4014
	Q4 1073–1358	191	3130
	Q5 ≥1359	193	4062
		$P = 0.49$	$P = 0.95$
Calcium, food sources only (mg/day)	Q1 ≤537	182	3959
	Q2 538–640	187	4006
	Q3 641–751	190	4032
	Q4 752–911	191	4041
	Q5 ≥912	201	4003
		$P = 0.009$	$P = 0.51$
Total vitamin A (IU/day)	Q1 <8136	183	3905
	Q2 8136–10704	186	3972
	Q3 10705–13636	194	4062
	Q4 13637–17895	200	4141
	Q5 >17895	190	3962
		$P = 0.08$	$P = 0.15$

Table 1 Continued

Dietary variable	Category[a] and range of intake	IGF-I (ng/ml)	IGFBP-3 (ng/ml)
Vitamin A, food sources only (IU/day)	Q1 <6998	178	3946
	Q2 6998–8821	187	3941
	Q3 8822–10909	199	4108
	Q4 10910–13963	195	4051
	Q5 >13963	192	3994
		$P = 0.02$	$P = 0.28$
α-Carotene (μg/day)	Q1 <381	185	3983
	Q2 381–549	191	3988
	Q3 550–738	198	4130
	Q4 739–1096	189	3956
	Q5 >1096	188	3983
		$P = 0.85$	$P = 0.87$
β-Carotene (μg/day)	Q1 <2592	180	3937
	Q2 2593–3530	192	3991
	Q3 3531–4476	198	4079
	Q4 4477–5907	187	4020
	Q5 >5907	194	4014
		$P = 0.15$	$P = 0.34$
β-Cryptoxanthin (μg/day)	Q1 <1261	180	3964
	Q2 1261–1760	186	3977
	Q3 1761–2298	197	4064
	Q4 2299–3170	197	4050
	Q5 >3170	192	3987
		$P = 0.03$	$P = 0.52$
Carotene (IU/day)	Q1 ≤5407	180	3918
	Q2 5408–7308	192	4041
	Q3 7309–9229	194	3973
	Q4 9230–12493	191	4119
	Q5 ≥12494	194	3990
		$P = 0.08$	$P = 0.27$
Lutein/Zeaxanthin (μg/day)	Q1 ≤1867	191	4018
	Q2 1868–2586	184	3969
	Q3 2587–3413	188	3950
	Q4 3414–4683	198	4118
	Q5 >4683	191	3986
		$P = 0.34$	$P = 0.66$
Lycopene (μg/day)	Q1 <5708	194	3973
	Q2 5709–7350	181	3831
	Q3 7351–9361	193	4051
	Q4 9362–11879	188	4036
	Q5 ≥11880	195	4149
		$P = 0.50$	$P = 0.003$
Total retinol (IU/day)	Q1 ≤1487	183	3991
	Q2 1488–2272	187	3975
	Q3 2273–3750	200	4033
	Q4 3751–6125	186	3969
	Q5 >6125	195	4073
		$P = 0.14$	$P = 0.40$
Retinol, food sources only (IU/day)	Q1 ≥1029	184	4025
	Q2 1030–1439	183	4008
	Q3 1440–1793	191	3949
	Q4 1794–2383	198	4096
	Q5 ≤2384	195	3963
		$P = 0.02$	$P = 0.84$
Total Vitamin E (mg/day)	Q1 ≤6.0	192	4045
	Q2 6.1–8.2	185	3878
	Q3 8.3–19.6	189	3983
	Q4 19.7–177.2	188	3988
	Q5 ≤177.3	197	4143
		$P = 0.36$	$P = 0.11$
Vitamin E, food sources only (mg/day)	Q1 ≤5.0	189	4044
	Q2 5.1–5.7	190	3996
	Q3 5.8–6.5	191	4016
	Q4 6.6–7.4	190	3974
	Q5 ≥7.5	191	4014
		$P = 0.83$	$P = 0.67$

Table 1 Continued

Dietary variable	Category[a] and range of intake	IGF-I (ng/ml)	IGFBP-3 (ng/ml)
Total Vitamin C (mg/day)	Q1 <126	187	3910
	Q2 127–177	196	4008
	Q3 178–250	186	4028
	Q4 251–484	196	4068
	Q5 ≥485	186	4027
		$P = 0.97$	$P = 0.13$
Vitamin C, food sources only	Q1 <97	180	3919
	Q2 97–126	197	4042
	Q3 127–153	188	3923
	Q4 154–189	193	4006
	Q5 >189	192	4151
		$P = 0.20$	$P = 0.03$
Total zinc (mg/day)	Q1 ≤9.6	180	4045
	Q2 9.7–10.9	190	3967
	Q3 11.0–12.6	194	3977
	Q4 12.7–18.1	188	3971
	Q5 ≥18.2	198	4081
		$P = 0.02$	$P = 0.68$
Zinc, food sources only (mg/day)	Q1 ≤9.2	178	3974
	Q2 9.3–10.1	190	4067
	Q3 10.2–11.0	195	4107
	Q4 11.1–12.1	197	3984
	Q5 ≥12.2	192	3905
		$P = 0.02$	$P = 0.24$
Caffeine (mg/day)	Q1 ≤70	197	4016
	Q2 71–156	183	3957
	Q3 157–261	190	4075
	Q4 262–385	195	4076
	Q5 ≥386	187	3916
		$P = 0.66$	$P = 0.70$

[a] Quintile unless otherwise noted.

supplements, had no association with either IGF-I or IGFBP-3. Although total vitamin C had little association with either IGF-I or IGFBP-3, vitamin C from food sources had a modest positive association with IGFBP-3. Zinc intake, whether from food sources only or including supplements, had a modest positive association with IGF-I. Caffeine intake was not associated with IGF-I or IGFBP-3.

Table 2 shows the multivariate associations of food groups and foods with IGF-I and IGFBP-3. Fruit, vegetable, and poultry intake had little association with either IGF-I or IGFBP-3 levels. Red meat intake had a borderline inverse association with IGFBP-3.

IGF-I levels were higher with greater intake of whole or skim milk, even after adjustment for multiple factors, but not with other dairy foods. In addition, higher intakes of cooked or cold breakfast cereal and pasta were associated with higher levels of IGF-I. Other grain-containing foods such as bread and rice did not have this association.

The positive association between energy intake and IGFs was confined to the leaner women. For the 611 women with a BMI <25, the multivariate adjusted mean levels of IGF-I (ng/ml) across increasing quintiles of energy intake were 178, 189, 193, 205, and 202 (P for the linear trend = 0.0007). The corresponding values for the 426 women with BMI ≥25 were 187, 179, 188, 188, and 184 ($P = 0.75$). However, the P for the interaction term (energy intake × overweight) was not statistically significant ($P = 0.11$).

Among dietary factors, the association of higher dairy intake with higher IGF-I levels was particularly consistent. This association was similar among women with and without obesity, a history of benign breast disease, or a family history of breast cancer or colon cancer. The positive association between dairy intake and IGF-I was seen in the 526 premenopausal women (lowest *versus* highest quartile of intake; P for the linear trend, 199 *versus* 226 ng/ml; $P = 0.001$), with little relationship in the 456 postmenopausal women (174 *versus* 174 ng/ml; $P = 0.46$). However, again, the P for the interaction term (dairy intake × menopausal status) was not statistically significant ($P = 0.54$). Results were similar for the comparison between premenopausal women and the 306 postmenopausal women not currently using HRT.

The inverse association between IGFBP-3 and saturated fat appeared to be confined to postmenopausal women only. The mean IGFBP-3 levels (ng/ml) across increasing quintiles of saturated fat intake among premenopausal women were 3844, 3855, 3823, 3952, 3872 (P for linear trend = 0.55). The corresponding IGFBP-3 levels for postmenopausal women, not using HRT, were 4493, 4255, 3970, 4115, and 4024 (P for linear trend = 0.02). The P for the interaction term (menopausal status × saturated fat intake) was 0.06. There was no significant interaction between menopausal status and other foods or nutrients.

Methods proposed to estimate the biologically active fraction of IGF-I did not substantially change the results. The molar ratio of IGF-I:IGFBP-3 has been proposed to reflect tissue bioactivity (29). We calculated it as (0.130 × IGF-I concentration in ng/ml) / (0.036 × IGFBP-3 concentration in ng/ml) for each of the factors in Tables 1 and 2. The positive association of total energy intake with IGF-I was attenuated with the

Table 2 Multivariate adjusted mean levels of IGF-I, IGFBP-3, and the molar ratio by quartile of food intake, and the *P* for the linear trend across categories

All dietary factors were adjusted for: energy intake, age, laboratory batch, menopausal status; use, type, and duration of hormone replacement; smoking, physical activity, parity, history of lactation and BMI. Categories the same as in the footnote to Table 1.

Dietary variable	Quartile and range of intake (servings/day)	IGF-I (ng/ml)	IGFBP-3 (ng/ml)
Fruit	Q1 <1.5	185	3987
	Q2 1.5–2.2	193	3981
	Q3 2.2–3.2	194	4035
	Q4 ≥3.2	188	4031
		$P = 0.58$	$P = 0.49$
Vegetables	Q1 ≤2.37	187	4051
	Q2 2.38–3.40	192	3972
	Q3 3.41–4.56	198	4028
	Q4 ≥4.57	184	3981
		$P = 0.90$	$P = 0.55$
Red meat	Q1 ≤0.6	195	4110
	Q2 0.6–0.9	189	4026
	Q3 0.9–1.2	186	3948
	Q4 >1.2	191	3949
		$P = 0.57$	$P = 0.04$
Poultry	Q1 <0.18	189	4017
	Q2 0.21–0.29	185	3933
	Q3 0.32–0.42	192	4024
	Q4 ≥0.43	194	4057
		$P = 0.25$	$P = 0.38$
Fish	Q1 <0.18	179	3917
	Q2 0.21–0.28	193	3970
	Q3 0.29–0.46	197	4016
	Q4 ≥0.46	193	4137
		$P = 0.008$	$P = 0.004$
Dairy	Q1 ≤1.1	188	4002
	Q2 1.2–1.7	182	3993
	Q3 1.7–2.5	190	4055
	Q4 ≥2.6	201	3983
		$P = 0.02$	$P = 0.96$
Milk	Q1 <0.3	183	3921
	Q2 0.3–0.7	189	4080
	Q3 0.7–1.6	188	3978
	Q4 ≥1.6	200	4035
		$P = 0.01$	$P = 0.37$
Ice cream	Q1 ≤0.03	190	4076
	Q2 0.07	191	4057
	Q3 0.10–0.14	190	3952
	Q4 >0.21	190	3917
		$P = 1.00$	$P = 0.02$
Yogurt	Q1 None	188	3970
	Q2 ≤0.03	187	4019
	Q3 0.07–0.14	192	4058
	Q4 >0.21	194	4009
		$P = 0.28$	$P = 0.43$
Cheese	Q1 <0.3	193	4041
	Q2 0.3–0.5	191	3957
	Q3 0.5–0.8	187	4092
	Q4 ≥0.8	190	3945
		$P = 0.53$	$P = 0.53$
Grains	Q1 ≤1.6	181	3915
	Q2 1.6–2.2	192	4088
	Q3 2.2–3.1	187	3998
	Q4 >3.1	201	4032
		$P = 0.02$	$P = 0.40$
Cereal	Q1 ≤0.1	171	3816
	Q2 0.2–0.4	196	4153
	Q3 0.5–0.7	199	4063
	Q4 ≥0.8	195	4008
		$P = 0.0002$	$P = 0.04$
Bread	Q1 ≤0.5	184	3960
	Q2 0.6–0.9	188	3989
	Q3 0.9–1.7	194	4050
	Q4 >1.7	195	4033
		$P = 0.08$	$P = 0.27$
Rice	Q1 ≤0.07	187	3966
	Q2 0.11	188	4022
	Q3 0.14–0.21	191	3997
	Q4 >0.21	195	4062
		$P = 0.18$	$P = 0.26$
Pasta	Q1 <0.07	179	3906
	Q2 0.10–0.14	190	3967
	Q3 0.21–0.25	194	3901
	Q4 ≥0.28	196	4144
		$P = 0.02$	$P = 0.002$

molar ratio (1st *versus* 5th quintile, 0.167 *versus* 0.173; $P = 0.09$).

We also calculated mean levels of IGF-I across categories of predictors, controlling for levels of IGFBP-3 (and other covariates) and likewise, levels of IGFBP-3 controlling for levels of IGF-I. The positive association of total energy with IGF-I was also attenuated when controlled for levels of IGFBP-3 (1st *versus* 5th quintile, 184 *versus* 192 ng/ml; $P = 0.07$). Other findings were essentially unchanged.

Discussion

In this cross-sectional analysis among >1000 women, we found associations between IGF-I and IGFBP-3 and several foods and nutrients. In particular, we found that total energy and protein intake were positively associated with plasma IGF-I levels. The association with protein intake was largely attributable to higher IGF-I levels among women who consumed higher amounts of milk. We had the ability to average two dietary assessments collected 3–4 years prior and near the time of blood collection, which should reduce random error in dietary measurement. However, we examined many foods and nutrients; associations could occur by chance and should be replicated in other studies.

Plasma IGF-I levels are decreased by malnutrition, in both animals and humans, and protein is particularly important for increasing IGF-I levels during refeeding (17, 30). However, few studies have been examined linking dietary components in relation to IGF levels in well-nourished humans. Kaklamani *et al.* (18) examined the associations of dietary data collected by a FFQ with plasma IGF-I and IGFBP-3 levels in 130 healthy Greek subjects between 30 and 84 years of age. They found that IGF-I levels were positively associated with intake of red meat and total fat and inversely associated with intake of carbohydrates. In addition, they found that levels of IGFBP-3 were inversely associated with intake of saturated fat (18).

Goodman-Gruen and Barrett-Connor (19) reported a positive association between alcohol use and IGF-I levels among men and women in Rancho Bernardo, and the linear relationship was statistically significant among men. We found a nonlinear association between alcohol intake and IGF-I that was somewhat positive for low intakes but inverse for the highest category of intake. However, the distribution of alcohol intake in the Rancho Bernardo study, which was conducted in an older population (mean age of the women, 77 years), overlapped only the low end of the intake distribution in our study. Therefore, the results of the two studies were compatible. We also found a positive association between alcohol intake and IGFBP-3 levels. The significance of this finding is not clear, but it would tend to make bioavailable levels of IGF-I even lower in the highest category of alcohol intake.

We found a positive association between IGF-I levels and intake of total energy, after controlling for BMI and physical activity, with an 8% increase between the lowest and highest intake. We also found a positive association between protein intake and IGF-I levels (13% increase between extremes of intake). Turnover of epithelial cells requires substantial energy and protein and is increased by IGF-I (31). It is possible that at times of protein-calorie malnutrition, down-regulation of cell turnover may be advantageous to conserve protein and energy. Thus, the decline of IGF-I levels in the setting of malnourishment could serve a physiological purpose. On the other hand, we speculate that there may be a tendency in some individuals for excess nutrition to be associated with elevation of IGF-I levels and increased cellular turnover rate that over prolonged periods of time could lead to increased cancer risk. In an animal carcinogenesis model, supplementation of IGF-I attenuated the protective effect that dietary restriction had on tumor incidence (32). However, in this cross-sectional study, we cannot distinguish which comes first: whether high intake of energy leads to higher IGF-I levels or whether having high IGF-I levels causes an individual to consume more energy.

Total energy intake has been consistently associated with cancer development in animals (33–36), although less consistently in humans (37–39). The positive association of IGF-I levels with intake of total energy in our study was somewhat stronger among the leanest women. This association could also be a reflection of residual confounding with physical activity (21). It may be important in the future to assess the association between energy intake and human cancer risk stratified by obesity.

The association between protein and IGF-I appeared to be limited to animal and not vegetable protein intake. Among animal protein-containing food groups, we found no association with red meat or poultry intake and a moderate positive association with fish intake. The relationship between protein or animal protein intake and IGF-I was not completely attenuated when controlled for dairy intake. However, the lack of association between IGF-I and other sources of animal protein makes dairy food the most likely cause of the relationship between IGF-I and protein.

Devine *et al.* (20) reported that zinc intake, as measured by weighed diet records, was positively associated with IGF-I levels in >100 postmenopausal women enrolled in a study of calcium supplementation for bone density. Although the major source of zinc in the diet is animal products, in their study this association persisted after control for protein intake. The authors postulated that zinc is necessary for hepatic *IGF-I* gene synthesis (20). We also found that intake of zinc, whether from food sources only or including supplements, was associated with higher levels of IGF-I.

Among other micronutrients, we found a positive association with calcium from foods but not from total calcium including supplements. Likewise, we found a stronger association with vitamin A from foods, retinol from foods, and vitamin D from foods than from total vitamin A, retinol, and vitamin D including supplements. These findings cast doubt on calcium, vitamin A, retinol, and vitamin D being a cause of higher IGF-I levels. In fact, in models containing both dairy intake and calcium, vitamin A, retinol, or vitamin D from foods, the association with IGF-I is attenuated, except for vitamin A from foods, where the association is approximately the same. In 1990, the largest (nonsupplement) dietary source of calcium and vitamin D was dairy food, and dairy foods were among the highest contributors to the intake of vitamin A and retinol from foods as well. The positive association we observed between lycopene intake and IGFBP-3 is consistent with the inverse association of lycopene with prostate cancer risk (40).

Our most consistent dietary finding was the positive association of IGF-I levels with total dairy and milk intake. This association is the most logical explanation for our finding a stronger association with calcium from foods but not total calcium including supplements, and the similar results with vitamin A and retinol. Supporting this finding was a positive association with grain-containing foods, most strongly with breakfast cereal. We also found no association with dietary fiber. In our data, the correlation between milk intake and cereal intake was 0.29. It is possible that these findings are attributable to the joint consumption of cereal and milk.

Dairy foods other than milk (ice cream, yogurt, and

cheese) did not have an association with IGF-I. It is possible that either IGF-I in cows' milk or a substance in milk that stimulates endogenous production of IGF-I is inactivated during the processing of milk to ice cream, yogurt, or cheese. There was an inverse association between ice cream intake and levels of IGFBP-3. This may have to do with the saturated fat content of ice cream. However, cheese also has a high saturated fat content, and no such association was seen with cheese.

Two other studies have supported an effect of milk intake on IGF-I levels. A randomized trial of 204 men and women where the intervention was to encourage consumption of three servings/day of nonfat milk to affect bone remodeling found that the 101 subjects in the intervention group had a statistically significant 10% average increase in serum IGF-I levels, whereas the control group had no change in levels (41). In addition, Ma *et al.* (42) observed a positive association between intake of dairy food and IGF-I levels among 318 men enrolled in the Physicians' Health Study.

IGF-I occurs naturally in cows' milk, but orally ingested IGF-I has been thought not to be absorbed. Supporting this conclusion is an unpublished study done by the Monsanto Agricultural Co. and communicated to the Food and Drug Administration in 1989 (43). Because IGF-I mediates the actions of GH, this study was conducted in anticipation of the Food and Drug Administration's approval of the use of rBGH in dairy cows to improve milk production. In this study, rats were orally administered high doses of recombinant IGF-I. No changes were seen in the serum IGF-I levels, blood and urine chemistries, and organ weights of the treated rats, as were seen in rats administered IGF-I or GH parenterally. Thus, it was considered unlikely to have any effect when ingested orally by humans. However, a recent study has reported that radioactively labeled IGF-I in milk fed to neonatal rats could be detected in low levels in their systemic circulation (44). In addition, neonatal rats fed excess amounts of IGF-I had increased growth of specific organs (45).

It is uncertain whether results of absorption studies in neonatal rats are relevant to the adult humans in the present study. In any case, the Food and Drug Administration's approval of the use of rBGH for dairy cows did not occur until 1994, and thus, the blood samples in the present study, collected in 1989 and 1990, were not affected by rBGH. However, bovine IGF-I (which is identical to human IGF-I), normally occurs in the milk of untreated cows, which is identical to human IGF-I and is increased in rBGH-treated cows (43). The possibilities that IGF-I in milk could be absorbed by the human gut, or that a component of milk, a food ideally suited for the growth of the infant mammal, may stimulate endogenous production of IGF-I, deserve investigation.

The primary limitation of this study is its cross-sectional nature; there is no way to know whether factors associated with IGF levels determine those levels or are in fact determined by them. Because subjects were sampled to increase the range of fat and alcohol intake, the distribution of dietary factors in the study population does not necessarily reflect the distribution of these factors in the underlying population. The strengths of this study include its large size and the extensive information on diet and lifestyle factors collected over 25 years.

We examined many dietary factors and found associations with several. However, the clearest findings in this study of healthy women were a positive association between intake of total energy and milk and circulating IGF-I levels. The milk association is supported by several pieces of evidence. This includes positive associations between IGF-I and nutrients from food sources but not supplements where the most frequent food source of those nutrients is milk. The milk association is consistent with similar findings in a cross-sectional study of men (42) and a randomized trial of increasing milk intake (41). These results raise the possibility that milk consumption could influence cancer risk by a mechanism involving IGF-I. In fact, positive associations between milk intake and risk of prostate cancer have been reported (46–51). In the NHS, one or more servings of milk/day was associated with a higher risk of serous ovarian cancer (relative risk, 1.66; 95% confidence interval, 1.10–2.51) compared with three or fewer servings/month (52).

In conclusion, we found that women consuming higher levels of alcohol and lower levels of saturated fat had higher levels of IGFBP-3. In addition, those consuming higher levels of protein, energy, and milk had higher circulating IGF-I levels. These results raise the possibility that cancer risk could be reduced by dietary manipulation of IGF levels.

References

1. Pollak, M. Insulin-like growth factor physiology and cancer risk. Eur. J. Cancer, *36:* 1224–1228, 2000.

2. Harrela, M., Koistinen, H., Kaprio, J., Lehtovirta, M., Tuomilehto, J., Eriksson, J., Toivanen, L., Koskenvuo, M., Leinonen, P., Koistinen, R., and Seppala, M. Genetic and environmental components of interindividual variation in circulating levels of IGF-I. IGF-II, IGFBP-1, and IGFBP-3. J. Clin. Investig., *98:* 2612–2615, 1996.

3. Boonen, S., Mohan, S., Dequeker, J., Aerssens, J., Vanderschueren, D., Verbeke, G., Broos, P., Bouillon, R., and Baylink, D. J. Down-regulation of the serum stimulatory components of the insulin-like growth factor (IGF) system (IGF-I, IGF-II, IGF binding protein [BP]-3, and IGFBP-5) in age-related (type II) femoral neck osteoporosis. J. Bone Miner. Res., *14:* 2150–2158, 1999.

4. Janssen, J. A., Stolk, R. P., Pols, H. A., Grobbee, D. E., and Lamberts, S. W. Serum total IGF-I, free IGF-I, and IGFB-1 levels in an elderly population: relation to cardiovascular risk factors and disease [published erratum appears in Arterioscler. Thromb. Vasc. Biol., *18:* 1197, 1998]. Arterioscler. Thromb. Vasc. Biol., *18:* 277–282, 1998.

5. Chan, J. M., Stampfer, M. J., Giovannucci, E., Gann, P. H., Ma, J., Wilkinson, P., Hennekens, C. H., and Pollak, M. Plasma insulin-like growth factor-I and prostate cancer risk: a prospective study. Science (Wash. DC), *279:* 563–566, 1998.

6. Wolk, A., Mantzoros, C. S., Andersson, S-O., Bergström, R., Signorello, L. B., Lagiou, P., Adami, H-O., and Trichopoulos, D. Insulin-like growth factor 1 and prostate cancer risk: a population-based, case-control study. J. Natl. Cancer Inst., *90:* 911–915, 1998.

7. Harman, S., Metter, E., Blackman, M., Landis, P., and Carter, H. Serum levels of insulin-like growth factor 1 (IGF-I), IGF-II, IGF-binding protein-3, and prostate-specific antigen as predictors of clinical prostate cancer. J. Clin. Endocrinol. Metab., *85:* 4258–4265, 2000.

8. Ma, J., Pollak, M. N., Giovannucci, E., Chan, J. M., Tao, T., Hennekens, C. H., and Stampfer, M. J. Prospective study of colorectal cancer risk in men and plasma levels of insulin-like growth factor (IGF)-1 and IGF-binding protein-3. J. Natl. Cancer Inst., *91:* 620–625, 1999.

9. Giovannucci, E., Pollak, M. N., Platz, E. A., Willett, W. C., Stampfer, M. J., Majeed, N., Colditz, G. A., Speizer, F. E., and Hankinson, S. E. A prospective study of plasma insulin-like growth factor-1 and binding protein-3 and risk of colorectal neoplasia in women. Cancer Epidemiol. Biomark. Prev., *9:* 345–349, 2000.

10. Renehan, A. G., Painter, J. E., O'Halloran, D., Atkin, W. S., Potten, C. S., O'Dwyer, S. T., and Shalet, S. M. Circulating insulin-like growth factor II and colorectal adenomas. J. Clin. Endocrinol. Metab., *85:* 3402–3408, 2000.

11. Manousos, O., Souglakos, J., Bosetti, C., Tzonou, A., Chatzidakis, V., Trichopoulos, D., Adami, H. O., and Mantzoros, C. IGF-I and IGF-II in relation to colorectal cancer. Int. J. Cancer, *83:* 15–17, 1999.

12. Yu, H., Spitz, M. R., Mistry, J., Gu, J., Hong, W. K., and Wu, X. Plasma levels of insulin-like growth factor-I and lung cancer risk: a case-control analysis. J. Natl. Cancer Inst., *91:* 151–156, 1999.

13. Hankinson, S. E., Willett, W. C., Colditz, G. A., Hunter, D. J., Michaud, D. S., Deroo, B., Rosner, B., Speizer, F. E., and Pollak, M. Circulating concentrations of insulin-like growth factor-I and risk of breast cancer. Lancet, *351:* 1393–1396, 1998.

14. Bruning, P. F., Van Doorn, J., and Bonfrer, J. M. G. Insulin-like growth-factor-binding protein 3 is decreased in early-stage operable premenopausal breast cancer. Int. J. Cancer, *62:* 266–270, 1995.

15. Toniolo, P., Bruning, P., Akhmedkhanov, A., Bronfrer, J., Koenig, K., Lukanova, A., Shore, R., and Zeleniuch-Jacquotte, A. Serum insulin-like growth factor-I and breast cancer. Int. J. Cancer, *88:* 828–832, 2000.

16. Yu, H., and Rohan, T. Role of the insulin-like growth factor family in cancer development and progression. J. Natl. Cancer Inst., *92:* 1472–1489, 2000.

17. Thissen, J., Ketelslegers, J., and Underwood, L. Nutritional regulation of the insulin-like growth factors. Endocr. Rev., *15:* 80–101, 1994.

18. Kaklamani, V. G., Linos, A., Kaklamani, E., Markaki, I., Koumantaki, Y., and Mantzoros, C. S. Dietary fat and carbohydrates are independently associated with circulating insulin-like growth factor 1 and insulin-like growth factor-binding protein 3 concentrations in healthy adults. J. Clin. Oncol., *17:* 3291–3298, 1999.

19. Goodman-Gruen, D., and Barrett-Connor, E. Epidemiology of insulin-like growth factor-I in elderly men and women. Am. J. Clin. Epidemiol., *145:* 970–976, 1997.

20. Devine, A., Rosen, C., Mohan, S., Baylink, D., and Prince, R. Effects of zinc and other nutritional factors on insulin-like growth factor I and insulin-like growth factor binding proteins in proteins in postmenopausal women. Am. J. Clin. Nutr., *68:* 200–206, 1998.

21. Holmes, M. D., Pollak, M., and Hankinson, S. E. Lifestyle correlates of insulin-like growth factor I and insulin-like growth factor binding protein 3. Cancer Epidemiol. Biomark. Prev., *11:* 2002.

22. Willett, W. C. Nutritional Epidemiology. New York: Oxford University Press, 1998, pp. 101–147.

23. Holmes, M. D., Spiegelman, D., Willett, W. C., Manson, J. E., Hunter, D. J., Barbieri, R. L., Colditz, G. A., and Hankinson, S. E. Dietary fat intake and endogenous sex steroid hormone levels in postmenopausal women. J. Clin. Oncol., *18:* 3668–3676, 2000.

24. Willett, W. C., Sampson, L., Stampfer, M. J., Rosner, B., Bain, C., Witschi, J., Hennekens, C. H., and Speizer, F. E. Reproducibility and validity of a semiquantitative food frequency questionnaire. Am. J. Epidemiol., *122:* 51–65, 1985.

25. Wolever, T. M. S., Jenkins, D. J., Jenkins, A. L., and Josse, R. G. The glycemic index: methodology and clinical implications. Am. J. Clin. Nutr., *54:* 846–854, 1991.

26. Liu, S., Manson, J. E., Stampfer, M. J., Holmes, M. D., Hu, F. B., Hankinson, S. E., and Willett, W. C. Dietary glycemic load assessed by food-frequency questionnaire in relation to plasma high-density-lipoprotein cholesterol and fasting plasma triacylglycerols in postmenopausal women. Am. J. Clin. Nutr., *73:* 560–566, 2001.

27. Liu, S., Willett, W. C., Stampfer, M. J., Hu, F. B., Franz, M., Sampson, L., Hennekens, C. H., and Manson, J. E. A prospective study of dietary glycemic load, carbohydrate intake, and risk of coronary heart disease in US women. Am. J. Clin. Nutr., *71:* 1455–1461, 2000.

28. Searle, S., Speed, F., and Milliken, G. Population marginal means in the linear model: an alternative to least square means. Am. Stat., *34:* 216–221, 1980.

29. Juul, A., Dalgaard, P., Blum, W., Bang, P., Hall, K., Michaelsen, K., Muller, J., and Skakkebaek, N. Serum Levels of insulin-like growth factor (IGF)-binding protein-3 (IGFBP-3) in healthy infants, children, and adolescents: the relation to IGF-I, IGF-II, IGFBP-1, IGFBP-2, age, sex, body mass index, and pubertal maturation. J. Clin. Endocrinol. Metab., *80:* 2534–2542, 1995.

30. Strauss, D., and CD. T. Effect of dietary protein deprivation on insulin-like growth factor (IGF)-I and -II, IGF binding protein-2, and serum albumin gene expression in rat. Endocrinology, *127:* 1849–1860, 1990.

31. Jones, J., and Clemmons, D. Insulin-like growth factors and their binding proteins-biological actions. Endocr. Rev., *16:* 3–34, 1995.

32. Dunn, S. E., Kari, F. W., French, J., Leininger, J. R., Travlos, G., Wilson, R., and Barrett, J. C. Dietary restriction reduces insulin-like growth factor I levels, which modulates apoptosis, cell proliferation, and tumor progression in p53-deficient mice. Cancer Res., *57:* 4667–4672, 1997.

33. Welsch, C. W. Relationship between dietary fat and experimental mammary tumorigenesis: a review and critique. Cancer Res., *52* (Suppl. 7)*:* 2040S–2048S, 1992.

34. Ip, C. Quantitative assessment of fat and calorie as risk factors in mammary carcinogenesis in an experimental model. *In:* C. J. Mettlin and K. Aoki (eds.), Recent Progress in Research on Nutrition and Cancer: Proceedings of a Workshop Sponsored by the International Union Against Cancer, held in Nagoya, Japan, November 1–3, 1989, pp. 107–117. New York: Wiley-Liss, Inc., 1990.

35. Boissonneault, G. A., Elson, C. E., and Pariza, M. W. Net energy effects of dietary fat on chemically induced mammary carcinogenesis in F344 rats. J. Natl. Cancer Inst., *76:* 335–338, 1986.

36. Tannenbaum, A. The dependence of the genesis of induced skin tumors on the caloric intake during different stages of carcinogenesis. Cancer Res., *4:* 673–677, 1944.

37. Holmes, M., Hunter, D., Colditz, G., Stampfer, M., Hankinson, S., Speizer, F., Rosner, B., and Willett, W. Association of dietary intake of fat and fatty acids with risk of breast cancer. J. Am. Med. Assoc., *281:* 914–920, 1999.

38. Giovannucci, E., and Willett, W. C. Dietary lipids and colon cancer. Princ. Pract. Oncol. Updates, *9:* 1–12, 1995.

39. Clinton, S. K., and Giovannucci, E. Diet, nutrition, and prostate cancer. Annu. Rev. Nutr., *18:* 413–440, 1998.

40. Giovannucci, E. Tomatoes, tomato-based products, lycopene, and cancer: review of the epidemiologic literature. J. Natl. Cancer Inst., *91:* 317–331, 1999.

41. Heaney, R., McCarron, D., Dawson-Hughes, B., Oparil, S., Berga, S., Stern, J., Barr, S., and Rosen, C. Dietary changes favorably affect bone remodeling in older adults. Am. Diet. Assoc., *99:* 1229–1233, 1999.

42. Ma, J., Giovannucci, E., Pollak, M., Chan, J., Gaziano, M., Willett, W., and Stampfer, M. Milk intake, circulating levels of IGF-1 and risk of colorectal cancer in men. J. Natl. Cancer Inst., *93:* 1330–1336, 2001.

43. Juskevich, J. C., and Guyer, C. G. Bovine growth hormone: human food safety evaluation [see comments]. Science (Wash. DC), *249:* 875–884, 1990.

44. Philipps, A. F., Dvorak, B., Anderson, G. G., Grille, J. G., Williams, C. S., and Koldovsky, O. Secretion of insulin-like growth factor-II into bile of rats of different ages. Biol. Neonate, *78:* 106–112, 2000.

45. Philipps, A. F., Anderson, G. G., Dvorak, B., Williams, C. S., Lake, M., Lebouton, A. V., and Koldovsky, O. Growth of artificially fed infant rats: effect of supplementation with insulin-like growth factor I. Am. J. Physiol., *272:* R1532–R1539, 1997.

46. Chan, J. M., Giovannucci, E., Andersson, S-O., Yuen, J., Adami, H-O., and Wolk, A. Dairy products, calcium, phosphorous, vitamin D, and risk of prostate cancer. Cancer Causes Control, *9:* 559–566, 1998.

47. Talamini, R., La Vecchia, C., Decarli, A., Negri, E., and Franceschi, S. Nutrition, social factors, and prostatic cancer in a Northern Italian population. Br. J. Cancer, *53:* 817–821, 1986.

48. Tzonou, A., Signorello, L. B., Lagiou, P., Wuu, J., Trichopoulos, D., and Trichopoulou, A. Diet and cancer of the prostate: a case-control study in Greece. Int. J. Cancer, *80:* 704–708, 1999.

49. La Vecchia, C., Negri, E., D'Avanzo, B., Franceschi, S., and Boyle, P. Dairy products and the risk of prostatic cancer. Oncology, *48:* 406–410, 1991.

50. Talamini, R., Franceschi, S., La Vecchia, C., Serraino, D., Barra, S., and Negri, E. Diet and prostatic cancer: a case-control study in northern Italy. Nutr. Cancer, *18:* 277–286, 1992.

51. Schuurman, A. G., van den Brandt, P. A., Dorant, E., and Goldbohm, R. A. Animal products, calcium and protein and prostate cancer risk in The Netherlands Cohort Study. Br. J. Cancer, *80:* 1107–1113, 1999.

52. Fairfield, K., Hunter, D., Colditz, G., Fuchs, C., Cramer, D., Speizer, F., Willett, W., and Hankinson, S. A prospective study of dietary lactose and ovarian cancer. J. Gen. Int. Med., *15:* 205–206, 2000.