Null Results in Brief

No Association between Dietary Phytoestrogens and Risk of Premenopausal Breast Cancer in a French Cohort Study

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Introduction

Phytoestrogens, plant food components with estrogen-like biological properties, are hypothesized to contribute to the 5-fold lower breast cancer incidence in Asian compared with Western countries (1). Isoflavones comprise the phytoestrogens most abundant in soy, the traditional staple food in Asia, and a recent meta-analysis concluded that there was a slight reduction in premenopausal breast cancer risk with higher soy consumption (1). Because consumption of soy and isoflavones is typically low in Western countries, lignans and their derived metabolites, the enterolignans, might be more relevant for breast cancer prevention in these populations (2). Further large prospective studies of phytoestrogens in breast cancer are needed in Western populations to test this hypothesis. We thus examined the association between the usual dietary intake of phytoestrogens and the risk of premenopausal invasive breast cancer in a large French cohort.

Materials and Methods

E3N (Etude Epidémiologique auprès de femmes de la Mutuelle Générale de l'Education Nationale) is a large ongoing prospective cohort consisting of 98,995 French women born between 1925 and 1950, subscribing to the health insurance plan for public education system employees, and who voluntarily enrolled in 1990-1991 (3). After a baseline questionnaire, follow-up questionnaires have been sent biennially to ascertain occurrence of diseases and to update menopausal status and exposure factors. Usual diet over the previous year was assessed using a validated 208-item diet history questionnaire administered between 1993 and 1995 and available for 74,524 participants (4). We estimated daily phytoestrogen intake using a food composition table updated for four isoflavones (genistein, daidzein, formononetin, and biochanin A), one coumestan (coumestrol), four plant lignans (pinoresinol, lariciresinol, secoisolariciresinol, and matairesinol), and two enterolignans (enterodiol and enterolactone). Dietary intake of total phytoestrogens was computed as the sum of isoflavones, coumestrol, and plant lignans.

All premenopausal women with dietary data, without a history of cancer (except for skin basal cell carcinoma or breast lobular carcinoma *in situ*), and who were not consuming soy dietary supplements were included in the present analysis (n = 26,868). Participants contributed person-years of follow-up starting from the date they had completed the dietary questionnaire to the date of diagnosis of premenopausal invasive breast cancer as first primary cancer (for the cases), date of diagnosis of another cancer, date of menopause, date of death, or July 2002, whichever came first. We calculated multivariate relative risks and their two-sided 95% confidence intervals in Cox proportional hazards regression models for quartiles of phytoestrogen intake, adjusting for potential confounding variables as listed in the footnotes to Table 1. We also conducted analyses stratified on the joint estrogen receptor (ER) and progesterone receptor (PR) status of the tumors.

Results

During 117,652 person-years of follow-up (median duration, 4.2 years), 402 cases of invasive breast cancer were diagnosed among 26,868 premenopausal women (mean age, 47 years at baseline). Median dietary intake of total phytoestrogens was 1,101 μg/d, mostly consisting of plant lignans (97%).

Premenopausal breast cancer risk was not related to isoflavone, coumestrol, plant lignan, or enterolignan intakes (Table 1). Nor was any association observed with individual intakes of genistein, daidzein, formononetin, biochanin A, coumestrol, pinoresinol, lariciresinol, secoisolariciresinol, matairesinol, enterodiol, or enterolactone (data not shown).

Most (80%) of the 322 breast cancer cases with known receptor status were positive for both ER and PR [191 (59%) ER^+PR^+, compared with 51 (16%) ER^-PR^-, 44 (14%) ER^+PR^-, and 36 (11%) ER^-PR^+]. When we stratified the analysis on the joint ER/PR status, no association was found (data not shown).

Conclusions

In this prospective study, we found no evidence of an association between dietary intake of phytoestrogens and risk

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Table 1. Multivariate relative risks and their 95% confidence intervals for invasive breast cancer according to quartiles of daily intake of dietary phytoestrogens among 26,868 premenopausal women in the E3N cohort

Dietary intake*	Range†, μg/d	Cases (N = 402)	Person-years (117,652)	Adjusted RR (95% CI)‡	P_{trend}§
Total isoflavones	1-22	107	29,799	1.00	
	22-28	78	29,549	0.73 (0.54-0.98)	
	29-35	110	29,097	1.03 (0.79-1.34)	
	36-112	107	29,207	1.00 (0.76-1.31)	0.48
Coumestrol	0	63	20,659	1.00	
	0.00-0.02	126	32,317	1.32 (0.97-1.80)	
	0.03-0.05	97	31,944	1.02 (0.74-1.40)	
	0.06-0.60	116	32,732	1.22 (0.89-1.66)	0.68
Total plant lignans	41-843	101	30,918	1.00	
	844-1,070	106	29,509	1.06 (0.81-1.40)	
	1,071-1,356	91	28,843	0.93 (0.70-1.23)	
	1,357-4,611	104	28,381	1.07 (0.81-1.41)	0.80
Total enterolignans‖	168-902	107	30,320	1.00	
	903-1,075	105	29,644	0.99 (0.75-1.30)	
	1,076-1,288	90	28,785	0.86 (0.65-1.14)	
	1,289-3,361	100	28,903	0.94 (0.71-1.24)	0.53

Abbreviations: RR, relative risk; 95% CI, 95% confidence interval.

*Total isoflavone intake was computed as the sum of individual isoflavones (genistein, daidzein, formononetin, and biochanin-A), total plant lignans as the sum of individual plant lignans (pinoresinol, lariciresinol, secoisolariciresinol, and matairesinol), and total enterolignans as the sum of individual enterolignans (enterodiol and enterolactone). All were adjusted for energy intake from food (excluding energy from alcohol from total energy intake) by the residual method (24).

† The range for each energy-adjusted phytoestrogen quartile was calculated by adding the residual range to the predicted phytoestrogen intake for the mean caloric intake from food (2,149 kcal) for the whole population according to the regression model. Specifically for coumestrol, we computed the lowest category with null values (18%) and higher categories from tertiles of non-null values.

‡ Multivariate RRs and 95% confidence intervals calculated by Cox proportional hazards regression models using age as the time scale and adjusted for years of education (≤12, 13-16, ≥15), height (as continuous variable), body mass index category (as a time-dependent variable according to the height at baseline and the weight at the start of each follow-up interval), age at menarche (<13, 13-14, ≥15 years), personal history of benign breast disease (including fibrocystic breast disease, mastosis, and adenoma) or lobular carcinoma *in situ* (yes or no), family history of breast cancer in first- or second-degree relatives (yes or no), lifetime use of oral contraceptive (yes or no), age at first full-term pregnancy (FFTP) and parity (nulliparous, age at FFTP <30 years and 1-2 children, age at FFTP <30 years and ≥3 children, or age at FFTP ≥30 years whatever the number of children), geographic area, alcohol consumption (as continuous variable), and dietary energy intake from food.

§ Test for linear trend using median values in each quartile as an ordinal variable.

‖ To account for the lack of data for some enterolignan values in the food composition table, we computed enterolignan values from lignan content using conversion factors obtained *in vitro* (25).

of premenopausal invasive breast cancer, either overall or by ER/PR status.

Dietary isoflavone (<120 μg/d) and coumestrol (<1 μg/d) intakes were close to those reported for other Western populations consuming little or no soy (2, 5, 6). In comparison, mean isoflavone intakes were ~15 mg/d in European soy consumers (7) and varied between 5 and 45 mg/d in Asian populations (8, 9). Our dietary questionnaire did not cover soy foods, but the proportion of soy consumers is marginal in France, with only 1% to 3% women (10, 11). Chronic intake of >1 g/d soy protein, corresponding to >3 mg/d isoflavones (9), was recently suggested for reducing premenopausal breast cancer risk (1). In line with this hypothesis, isoflavone intake levels in this study may have been too low to reveal an association. The use of soy supplements in Western premenopausal women would enable attaining Asian isoflavone intakes; however, this is not recommended for reducing breast cancer risk insofar as its safety has not yet been shown (1).

In our study, plant lignan intakes were 30 times higher than isoflavone intakes. Our study evaluated the contributions of two major lignans newly identified (12) and reported higher lignan intakes compared with other authors. Whereas previous studies may have been limited by too low lignan intakes, inverse associations were shown for premenopausal breast cancer of certain CYP17 genotypes and ER^- status (13-15) and of ER^+ and PR^+ status (16); conversely, two dietary reports found no association, just as we did (17, 18). Limited population size and follow-up duration might explain the absence of an association in our study despite a larger range of lignan intakes. However, we had adequate statistical power (80%, with 5% two-sided significance) to detect a relative risk of ≥1.43 for the higher quartile of lignan intakes, with the sample size available. The lack of trend across quartiles of lignan intakes and our finding of inverse associations at similar levels in postmenopausal women[3] suggest no real association with lignans in this study.

Enterolignans are metabolized from ingested dietary lignans in the gut and are the bioactive compounds absorbed. The absence of an association in our study does not confirm results of a dietary case-control study with similar intake levels that showed a reduced risk of premenopausal breast cancer with higher enterolignan intakes (16). Three prospective (19-21) and one case-control (22) biomarker studies of enterolignans in premenopausal breast cancer showed inconsistent results. As our study had sufficient statistical power to detect a substantial risk reduction with enterolignan intakes (80% power to detect a relative risk of ≤0.65), it suggests that enterolignans are unlikely to be associated with risk.

In summary, the absence of an association in the present study probably indicates that there are no effects of low isoflavone and high lignan levels in premenopausal breast cancer. However, a balanced diet rich in plant foods remains recommended for Western premenopausal women, as a healthy diet is likely to be beneficial over the long term (23).

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