

Soy intake and blood cholesterol concentrations: a cross-sectional study of 1033 pre- and postmenopausal women in the Oxford arm of the European Prospective Investigation into Cancer and Nutrition¹⁻³

Magdalena S Rosell, Paul N Appleby, Elizabeth A Spencer, and Timothy J Key

ABSTRACT

Background: Clinical trials have suggested that the intake of soy protein reduces blood cholesterol. Few studies have explored this relation in subjects who consume soy as part of their regular diet.

Objective: In this study, we investigated whether blood cholesterol concentrations are related to the intake of soyfoods in a cohort comprising subjects with a wide variation in soy intake.

Design: This cross-sectional study included 1033 pre- and postmenopausal women selected from the Oxford arm of the European Prospective Investigation into Cancer and Nutrition. The sample included 361 nonvegetarians, 570 vegetarians, and 102 vegans. Their dietary intake was assessed by using a food-frequency questionnaire. Anthropometric data, medical history, and lifestyle information were obtained with the use of a questionnaire, blood samples were obtained, and plasma total, LDL-, and HDL-cholesterol concentrations were measured.

Results: Soy-protein intake was inversely associated with total and LDL-cholesterol concentrations and with the ratio of total to HDL cholesterol but not with HDL-cholesterol concentrations. Mean plasma LDL-cholesterol concentrations in women with a soy-protein intake ≥ 6 g/d was 12.4% lower than that in women who consumed <0.5 g/d ($P < 0.001$).

Conclusion: Moderate intakes of soyfoods as part of a regular diet are associated with favorable blood cholesterol concentrations. *Am J Clin Nutr* 2004;80:1391-6.

KEY WORDS Soy intake, blood cholesterol, cross-sectional study, vegetarians

INTRODUCTION

A high blood concentration of LDL cholesterol is an established risk factor for cardiovascular disease (1). After allowance for measurement error, a reduction in cholesterol concentration of 1 mmol/L is associated with a $\geq 21\%$ reduction in coronary artery disease risk (2). Although women have a lower risk than do men, the lifetime risk of coronary artery disease in women is still 1 in 3 (3). The delayed development of coronary artery disease in women compared with men may be partly explained by a favorable lipoprotein profile in premenopausal women. This profile changes after menopause, when total and LDL-cholesterol concentrations increase (4).

Blood cholesterol concentrations are influenced by diet. The relation between a high intake of saturated fats and increased blood cholesterol concentrations is well documented (5). Many

clinical trials have also suggested that increasing the intake of soy has favorable effects on blood cholesterol. A meta-analysis by Anderson et al (6) from 1995 indicated that a daily intake of 47 g soy protein is associated with a 12.9% reduction in LDL cholesterol. Since then, several trials have been conducted to investigate whether the hypocholesterolemic effect of soy is attributable to the soy protein or the high content of the phytoestrogenic isoflavones in soy. Some studies showed a specific effect of isoflavones on LDL or HDL cholesterol (7-11), whereas others did not (12-18). In a recent meta-analysis, beneficial effects of soy protein on LDL and HDL cholesterol were confirmed; however, no independent effect of soy-associated isoflavones was found (19).

The amounts of soy used in clinical trials have usually been large, typically within the range of 20-60 g soy protein/d. In contrast, few studies have investigated whether blood cholesterol concentrations are related to regular intakes of soy in amounts typical of people's habitual diet. We investigated the relation between the intake of soy protein and plasma cholesterol concentrations in 1033 pre- and postmenopausal women who participated in the Oxford arm of the European Prospective Investigation into Cancer and Nutrition (EPIC) study. The study sample includes a large proportion of vegetarians and vegans (65%) and involves subjects who consume diets with a wide range of soy content.

SUBJECTS AND METHODS

Subjects

The EPIC-Oxford cohort consists of 65 500 participants aged ≥ 20 y who were living in the United Kingdom between 1993 and 1999. The aim was to recruit participants with a wide range of diets by targeting vegetarians and vegans as well as the general

¹ From the Cancer Research UK Epidemiology Unit, University of Oxford, Oxford, United Kingdom.

² EPIC-Oxford is supported by Cancer Research UK, the Medical Research Council, and the European Commission under the Europe Against Cancer Programme. MSR was supported by a grant from the Swedish Council for Working Life and Social Research.

³ Reprints not available. Address correspondence to MS Rosell, Cancer Research UK Epidemiology Unit, University of Oxford, Gibson Building, Radcliffe Infirmary, Oxford OX2 6HE, United Kingdom. E-mail: magdalena.rosell@cancer.org.uk.

Received April 20, 2004.

Accepted for publication June 15, 2004.

UK population. The study protocol was approved by a Multi-Centre Research Ethics Committee. Participants were recruited through collaborating general practitioners, vegetarian and vegan societies, vegetarian and health food magazines, and friends or relatives of other participants (20). The proportions of vegetarians and vegans in the EPIC-Oxford cohort were 28.8% and 4.0%, respectively. Participants completed a detailed questionnaire that included questions on medical history, diet, and lifestyle. Diet during the previous 12 mo was assessed by using a 130-item food-frequency questionnaire (FFQ) (20). Blood samples were obtained from 19 700 volunteers.

The present study is based on a sample of 1091 pre- and postmenopausal women who were recruited into the EPIC-Oxford study during 1994 and 1995 and who gave a blood sample (21). The women were selected as being either clearly premenopausal (having ≥ 10 menstrual periods in the past 12 mo and a most recent menstrual cycle of < 40 d) or clearly postmenopausal (having had no menstrual periods in the past 12 mo) and as representing a wide range of dietary habits (meat-eaters, vegetarians, and vegans). The subjects were not taking exogenous sex hormones at recruitment, had not undergone hysterectomy, and had not been diagnosed with cancer of any type. For the present study, we excluded 3 women who did not have data on plasma lipids, 17 women with unreliable nutrient intake data ($> 20\%$ of the FFQ questions unanswered or energy intake of < 2.1 or > 14.7 MJ/d), 7 women who were taking hypolipidemic agents at blood draw or at recruitment, 1 woman with unknown smoking habits, and 30 women with unknown body mass index (BMI; in kg/m^2), which left 1033 women for the analysis.

Assessment of soy-protein intake

Two of the questions in the FFQ were directly related to the consumption of soy: 1) what type of milk the subjects consumed most often (soymilk was one of the options) and the amount consumed each day, including milk with tea, coffee, and cereals (0, 0.25, 0.50, 0.75, 1, or > 1 pint; 1 pint = 568 mL); and 2) the frequency of consumption of soy foods, such as tofu, soy meat, textured vegetable protein, and veggie burgers (never or $< 1/\text{mo}$, 1–3/mo, 1/wk, 5–6/wk, 1/d, 2–3/d, 4–5/d, or $\geq 6/\text{d}$). In addition, some subjects indicated that they consumed soy-based substitutes for dairy foods (cream, yogurt, dairy desserts, milk puddings, ice cream, cheese, and cottage cheese), and these intakes were also included in the calculation of soy-protein intake. The daily intake of soy protein was estimated from the reported consumption of soymilk, solid soy foods, and, where applicable, soy “dairy” foods as listed above by using British food-composition tables (22–24). The subjects were categorized into 4 groups of soy-protein intake for the analysis: < 0.5 , 0.5–2.9, 3.0–5.9, and ≥ 6.0 g/d. The cutoffs were chosen to give 4 distinct categories of soy-protein intake, each containing a sizeable number of subjects. The same questions in the FFQ were also used to estimate the intake of isoflavones from soyfoods by using data from the US Department of Agriculture–Iowa State University database (25). The estimated intakes of soy protein and isoflavones were highly correlated ($r = 0.98$), and therefore we used only soy-protein intake as the exposure variable in the analyses.

Measurement of plasma lipids

Blood samples (30 mL) were obtained from the subjects on average 5 mo after they completed the questionnaire. The blood

samples were sent via mail to the central laboratory and separated into plasma aliquots, which were frozen at -70°C and stored until analysis. Plasma lipid concentrations were measured at the Clinical Biochemistry Laboratory, Addenbrooke's Hospital, Cambridge, United Kingdom. Total cholesterol and triacylglycerol concentrations were measured by using automated enzymatic procedures with reagents supplied by Bayer (Newbury, United Kingdom), and HDL-cholesterol concentrations were measured by using a similar method after pretreatment with a precipitant supplied by Boehringer Mannheim (Lewes, United Kingdom). LDL cholesterol was calculated according to the Friedewald formula as total cholesterol minus triacylglycerols/2.19 minus HDL cholesterol (26). The interassay CVs were 1.4% and 1.3% at a total cholesterol concentration of 2.6 mmol/L and an HDL-cholesterol concentration of 1.3 mmol/L, respectively.

Other variables

The FFQ was also used to estimate the intake of fats, protein, carbohydrate, cholesterol, fiber (nonstarch polysaccharides), alcohol, and energy by using British food-composition tables (20). Information on age, height, weight, alcohol intake, smoking habits, physical activity, use of hormone replacement therapy, menopausal status, illness, and medications was obtained from the questionnaire.

Statistics

Each of the plasma lipid concentrations (total, LDL, and HDL cholesterol) together with the ratio of total to HDL cholesterol were log-transformed to correct for positive skewness and to make the distributions more nearly normal. The association between each of these variables and soy-protein intake was investigated by using multiple linear regression with adjustment for age (as a continuous variable and including a quadratic term), nondietary factors, BMI, and selected nutrient intakes. The nondietary factors were menopausal status (categorized as pre- or postmenopausal), cigarette smoking (never, former, or current), alcohol consumption (< 1 , 1–7, 8–15, or ≥ 16 g ethanol/d), and previous use of hormone replacement therapy (never user or past user). BMI was categorized as < 20.0 , 20.0–22.4, 22.5–24.9, 25.0–27.4, 27.5–29.9, or ≥ 30 . Partial correlation coefficients were used to explore the association between nutrient intakes and plasma lipid concentrations, with adjustment for age and menopausal status. The nutrients selected for inclusion in the regression analysis were energy, saturated fatty acids (SFAs) as a percentage of energy, polyunsaturated fatty acids (PUFAs) as a percentage of energy, dietary cholesterol, and nonstarch polysaccharides (NSPs), each as a continuous variable. Diet group was characterized as nonvegetarian (women who ate meat and fish), vegetarian (women who did not eat meat or fish but ate dairy products and eggs), or vegan (women who did not eat any animal products).

Adjusted geometric mean lipid concentrations by soy-protein intake category (< 0.5 , 0.5–2.9, 3.0–5.9, and ≥ 6.0 g/d) together with their 95% CIs were calculated from the fitted values arising from the regression models. A test of trend was obtained by substituting the logarithm of soy-protein intake for the categorical soy-protein variable in the regression model. Subset analyses were performed according to menopausal status and diet group. All analyses were performed by using version 8.1 of the STATA statistical package (release 8.0; Stata Corporation, College Station, TX).



TABLE 1
Characteristics of the women by soy-protein intake category¹

Characteristic	Soy-protein intake (g/d)				Total	P ²
	<0.5 [n = 323 (31%)]	0.5–2.9 [n = 264 (26%)]	3.0–5.9 [n = 282 (27%)]	≥6.0 [n = 164 (16%)]		
Menopausal status						
Premenopausal [n (%)]	131 (41)	164 (62)	204 (72)	116 (71)	615 (60)	
Age (y)	38 (21–44) ³	37 (20–44)	36 (20–45)	34 (20–44)	37 (20–45)	<0.001
Postmenopausal [n (%)]	192 (59)	100 (38)	78 (28)	48 (29)	418 (40)	
Age (y)	64 (54–76)	63 (55–85)	64.5 (55–75)	62.5 (55–74)	64 (54–85)	0.231
Cigarette smoking [n (%)]						
Never	208 (64)	176 (67)	176 (62)	117 (71)	677 (66)	
Former	98 (30)	71 (27)	89 (32)	39 (24)	297 (29)	0.574
Current	17 (5)	17 (6)	17 (6)	8 (5)	59 (6)	
Alcohol intake (g ethanol/d)	5.8 ± 7.7 ⁴	6.2 ± 8.7	6.5 ± 9.5	5.8 ± 8.7	6.1 ± 8.6	0.708
Previous use of HRT [n (%)]	31 (10)	18 (7)	5 (2)	10 (6)	64 (6)	0.001
Diet group [n (%)]						
Nonvegetarian	242 (75)	78 (30)	30 (11)	11 (7)	361 (35)	
Vegetarian	80 (25)	184 (70)	227 (80)	79 (48)	570 (55)	<0.001
Vegan	1 (0)	2 (1)	25 (9)	74 (45)	102 (10)	
BMI (kg/m ²)	23.9 ± 4.1	22.8 ± 3.7	22.7 ± 3.5	22.0 ± 2.7	23.0 ± 3.7	<0.001
Daily nutrient intake						
Energy (MJ)	8.0 ± 2.2	7.9 ± 2.1	7.8 ± 2.1	7.6 ± 2.0	7.8 ± 2.1	0.279
Protein (% of energy)	16.4 ± 3.3	14.2 ± 2.7	14.3 ± 2.4	13.8 ± 2.1	14.9 ± 2.9	<0.001
Carbohydrate (% of energy)	49.9 ± 6.8	52.7 ± 6.7	52.3 ± 6.5	53.3 ± 6.5	51.8 ± 6.8	<0.001
Total fat (% of energy)	31.5 ± 6.6	30.8 ± 6.7	31.0 ± 6.4	30.7 ± 6.5	31.1 ± 6.6	0.429
SFAs (% of energy)	10.6 ± 3.7	9.4 ± 3.3	8.7 ± 3.1	6.0 ± 2.2	9.0 ± 3.6	<0.001
PUFAs (% of energy)	5.2 ± 2.1	5.5 ± 2.1	5.9 ± 2.4	8.1 ± 2.7	5.9 ± 2.5	<0.001
Dietary cholesterol (mg)	219 ± 111	159 ± 86	129 ± 80	54 ± 56	153 ± 105	<0.001
NSPs (g)	20.9 ± 8.2	22.3 ± 7.9	24.2 ± 8.2	25.9 ± 7.9	23.0 ± 8.2	<0.001

¹ HRT, hormone replacement therapy; SFAs, saturated fatty acids; PUFAs, polyunsaturated fatty acids; NSPs, nonstarch polysaccharides.

² P for differences between categories of soy-protein intake; differences in means were calculated by using ANOVA, and differences in proportions (cigarette smoking, previous use of HRT, and diet group) were calculated by using the chi-square test.

³ Median; range in parentheses (all such values).

⁴ $\bar{x} \pm SD$ (all such values).

RESULTS

Daily soy-protein intake ranged from 0 to nearly 30 g, with an overall mean of 3.0 g and a median of 1.0 g. The mean intakes in each of the 4 categories of soy-protein intake were 0.1, 0.8, 3.6, and 11.2 g/d, respectively. Five women (0.5%) had a soy-protein intake ≥ 25 g/d. The estimated intake of soy isoflavones ranged from 0 to >130 mg/d, with a mean of 10.2 mg/d and a median of 3.3 mg/d.

Characteristics of the women by soy-protein intake category are shown in **Table 1**. Sixty percent of the women were premenopausal at recruitment, and these women had a median age of 37 y; the remaining, postmenopausal women had a median age at recruitment of 64 y. The proportions of pre- and postmenopausal women differed between the soy intake categories ($P < 0.001$). A higher proportion of premenopausal women than of postmenopausal women consumed ≥ 6 g soy protein/d (19% compared with 11%). Only 6% of the women were current smokers, alcohol consumption was generally low (median daily intake of 2.5 g ethanol), and only 6% of the women reported past use of hormone replacement therapy.

On the basis of their responses to the FFQ, 361 (35%) of the women were nonvegetarian at recruitment, 570 (55%) were vegetarian, and 102 (10%) were vegan. Diet group was clearly associated with soy-protein intake. Seventy-five percent of the women in the lowest soy-protein intake category ate meat and

fish, whereas 93% of the women in the highest intake group were either vegetarian or vegan. This association was reflected in the mean values for BMI and daily nutrient intakes in the 4 soy-protein intake categories. Thus, the women who consumed ≥ 6 g soy protein/d were generally slimmer than were those who consumed <0.5 g soy protein/d, and the women who consumed ≥ 6 g soy protein/d had lower intakes of energy, protein, SFAs, and dietary cholesterol but higher intakes of carbohydrate, PUFAs, and NSPs. However, total fat intake was $\approx 31\%$ of energy in each soy-protein intake category.

Each of the logarithms of soy-protein intake, SFAs, PUFAs, dietary cholesterol, and NSPs were significantly correlated with LDL-cholesterol concentrations after adjustment for age and menopausal status; Pearson partial correlations were -0.22 , 0.14 , -0.13 , 0.19 , and -0.10 , respectively [$P < 0.001$ in each case except for NSPs ($P = 0.002$)]. Energy intake was not associated with LDL-cholesterol concentrations. In contrast, neither soy-protein intake nor any of the nutrients were significantly correlated with HDL-cholesterol concentrations.

Geometric mean plasma lipid concentrations by soy-protein intake category after adjustment for age, nondietary factors (menopausal status, cigarette smoking, alcohol consumption, and previous use of hormone replacement therapy), BMI, and intakes of energy, SFAs, PUFAs, dietary cholesterol, and NSPs are shown in **Table 2**. Results are shown for all women,

TABLE 2

Plasma lipid concentrations by soy-protein intake category after adjustment for age, nondietary factors, BMI, and selected nutrient intakes¹

Plasma lipid fraction and subset of women	Soy-protein intake (g/d)				<i>P</i> for trend
	<0.5	0.5–2.9	3.0–5.9	≥6.0	
Total cholesterol	<i>mmol/L</i>				
All	4.40 (4.31, 4.49)	4.33 (4.24, 4.42)	4.21 (4.13, 4.30)	4.07 (3.94, 4.20)	<0.001
Premenopausal	3.89 (3.77, 4.03)	3.91 (3.81, 4.02)	3.77 (3.68, 3.86)	3.68 (3.54, 3.83)	0.048
Postmenopausal	5.21 (5.08, 5.35)	5.00 (4.83, 5.17)	4.98 (4.79, 5.18)	4.70 (4.44, 4.98)	0.002
LDL cholesterol					
All	2.73 (2.64, 2.81)	2.66 (2.58, 2.74)	2.55 (2.47, 2.63)	2.39 (2.28, 2.50)	<0.001
Premenopausal	2.32 (2.21, 2.44)	2.32 (2.23, 2.42)	2.21 (2.13, 2.29)	2.10 (1.98, 2.22)	0.029
Postmenopausal	3.41 (3.29, 3.54)	3.21 (3.06, 3.36)	3.13 (2.96, 3.30)	2.89 (2.67, 3.13)	<0.001
HDL cholesterol					
All	1.26 (1.23, 1.30)	1.29 (1.25, 1.32)	1.26 (1.22, 1.29)	1.28 (1.23, 1.33)	0.924
Premenopausal	1.27 (1.22, 1.32)	1.29 (1.25, 1.33)	1.26 (1.23, 1.30)	1.30 (1.24, 1.36)	0.921
Postmenopausal	1.24 (1.20, 1.29)	1.27 (1.21, 1.33)	1.26 (1.19, 1.33)	1.28 (1.18, 1.38)	0.710
Total:HDL cholesterol					
All	3.48 (3.38, 3.59)	3.37 (3.27, 3.47)	3.36 (3.26, 3.45)	3.17 (3.04, 3.31)	0.006
Premenopausal	3.06 (2.94, 3.19)	3.03 (2.92, 3.13)	2.99 (2.90, 3.08)	2.84 (2.71, 2.98)	0.093
Postmenopausal	4.19 (4.03, 4.37)	3.94 (3.74, 4.14)	3.95 (3.72, 4.19)	3.68 (3.37, 4.02)	0.020

¹ All values are geometric \bar{x} ; 95% CI in parentheses. The nondietary factors were menopausal status, cigarette smoking, alcohol consumption, and previous use of hormone replacement therapy. The selected nutrients were energy, saturated fatty acids (as percentage of energy), polyunsaturated fatty acids (as percentage of energy), dietary cholesterol, and nonstarch polysaccharides. The test of trend was obtained by replacing the categorical soy-protein intake variable with the logarithm of soy-protein intake in the regression analysis.

premenopausal women, and postmenopausal women. *P* values for trend obtained by replacing the categorical soy-protein intake variable with the logarithm of soy-protein intake in the regression models are also shown in the table.

Soy-protein intake was significantly inversely associated with total cholesterol, LDL cholesterol, and the ratio of total to HDL cholesterol (*P* for trend < 0.01 for each). Compared with the women who consumed <0.5 g soy protein/d, those who consumed ≥6.0 g/d had a 7.5% lower mean total cholesterol concentration, a 12.4% lower mean LDL-cholesterol concentration, and a 9.0% lower mean ratio of total to HDL cholesterol (each *P* < 0.01). HDL cholesterol was not related to soy-protein intake.

The associations of soy-protein intake with total cholesterol, LDL cholesterol, and the ratio of total to HDL cholesterol were somewhat stronger in the postmenopausal women than in the premenopausal women although none of these differences was significant. Compared with the premenopausal women who consumed <0.5 g soy protein/d, the premenopausal women who consumed ≥6.0 g/d had a 5.4% lower mean total cholesterol concentration (*P* = 0.048), a 9.6% lower mean LDL-cholesterol concentration (*P* = 0.029), and a 7.3% lower mean ratio of total to HDL cholesterol (*P* = 0.093). The corresponding differences for the postmenopausal women were 9.9% (*P* = 0.002), 15.4% (*P* < 0.001), and 12.3% (*P* = 0.020), respectively.

The inverse associations of soy-protein intake with total cholesterol, LDL cholesterol, and the ratio of total to HDL cholesterol were slightly stronger for the nonvegetarian women than for either the vegetarian or the vegan women. However, there were no significant interactions between diet group and soy-protein intake in relation to plasma lipid concentrations (data not shown).

DISCUSSION

In this study of 1033 British women, plasma LDL-cholesterol concentrations in the women who reported a soy-protein intake

of ≥6 g/d were 12.4% lower than those in the women who reported an intake of <0.5 g/d. The association was slightly more pronounced in the postmenopausal women than in the premenopausal women. Similar results were found for total cholesterol and the ratio of total to HDL cholesterol. No association was seen between soy-protein intake and plasma HDL cholesterol.


An important methodologic concern in this study is the validity of the dietary data. The assessment of soy intake was mainly based on questions relating to the intake of soymilk and solid soy foods. We were unable to distinguish between different types of solid soy food (tofu, veggie burgers, etc), and therefore the amount of soy protein obtained from these foods was imprecise. However, foods that contain a large amount of soy protein are relatively limited, and because there were large differences in dietary intakes in this study, the questions in the FFQ can be reasonably assumed to have been sufficient for classifying subjects into different categories of soy intake. The reported intake of soy in the EPIC-Oxford cohort has also been shown to be strongly correlated with plasma concentrations of the isoflavones daidzein and genistein (27). Our results were adjusted for several potential dietary confounders. However, these variables are also subject to errors in measurement; in a validation study, the Spearman correlation coefficients between the FFQ used in the present study and 16-d weighed records for the intakes of energy and fat were 0.52 and 0.55, respectively (28). In the present study, an average intake of soy protein of 11 g/d was associated with a 12.4% reduction in LDL cholesterol. In the meta-analysis of clinical studies by Anderson et al (6), a similar reduction in LDL cholesterol (12.9%) was associated with a considerably higher intake of soy protein (47 g/d). This suggests that the beneficial effect of soy intake observed in the present study may be partly explained by other dietary factors for which we were unable to fully control.

Few studies have investigated the relation between blood cholesterol and intakes of soy that reflect people's habitual diet. In

these observational studies, soy was consumed in various forms (eg, as beans, tofu, and soymilk) that differ from the isolated or textured soy protein often used in clinical trials. Besides protein, the soybean contains fiber, fatty acids, and other nutrients and compounds that also may mediate beneficial health effects (29). In one Japanese study of 4838 subjects, the differences in total cholesterol concentrations between subjects in the highest and lowest quartiles of soy intake were -6% and -5% in men and women, respectively (30). In the Framingham Offspring Study, which included 939 postmenopausal women, no association between isoflavone intake and serum lipoproteins was observed; however, isoflavone intake was significantly inversely related to a measure of the metabolic syndrome (31). A study in California of 208 postmenopausal women showed a positive association of isoflavone intake with HDL cholesterol but no association with total or LDL cholesterol (32). However, isoflavone intakes in the 2 American studies were very low, which may explain why no associations between soy intake and total or LDL cholesterol were found. Our study differs from the previous studies because it involved subjects who lived in a Western country but had a wide range of soy intake. The average soy intake in our study was lower than that in the Japanese study but was considerably higher than that in the American studies. Our study also included both pre- and postmenopausal women, and the associations between soy intake and plasma cholesterol appeared to be slightly stronger among the postmenopausal women than among the premenopausal women. Although we do not know whether this reflects a true difference, the effect of diet on blood lipids may be greater in postmenopausal women because they have higher cholesterol concentrations than do premenopausal women, and the effects of soy have been shown to be more pronounced in subjects with hypercholesterolemia. In addition, in premenopausal women, hormonal influences on blood lipids may overshadow the possible effects of diet (33).

Blood cholesterol concentration is of interest because it is an important risk factor for cardiovascular disease. To our knowledge, only 2 studies have investigated whether soy intake is related to the risk of developing cardiovascular disease. One study including $>64\,000$ women in Shanghai, China, showed an inverse dose-response relation between soy intake and coronary artery disease risk; compared with women in the lowest quartile of soy-protein intake, those in the highest quartile had an adjusted relative risk of 0.25 (95% CI: 0.10, 0.63) (34). In a Japanese study, compared with men and women in the lowest quartile of intake of soy foods, men and women in the highest quartile had adjusted hazard ratios for cardiovascular disease mortality of 0.78 (95% CI: 0.55, 1.12) and 0.90 (95% CI: 0.63, 1.28), respectively (35). Further longitudinal data are evidently needed to establish the effect of soy intake on risks of morbidity and mortality.

A cholesterol-lowering effect of soy has been reported in many clinical trials, and the US Food and Drug Administration currently approves a health claim that consuming 25 g soy protein/d as part of a diet low in saturated fat and cholesterol may reduce the risk of heart disease (36). In our study, very few subjects consumed this amount of soy. However, reduced LDL-cholesterol concentrations were observed in subjects who consumed ≥ 6 g soy protein/d. The mean intake of soy protein in this group was 11 g/d, an intake that can be achieved by consuming, eg, 100 g tofu and 100 mL soymilk. The present study indicates that moderate intakes of soy foods as part of a regular diet are

associated with favorable blood cholesterol concentrations. This may be partly due to a biological effect of soy and partly due to the overall composition of diets with a high soy content. 

We thank all the participants in this study and the EPIC study staff at the Cancer Research UK Epidemiology Unit.

All authors were involved in discussing and interpreting the data and contributed to the writing of the manuscript. In addition, MSR wrote most parts of the manuscript. PNA performed the statistical analyses, wrote the Statistics and Results sections, and edited the manuscript. EAS edited the manuscript. TJK is the principal investigator of the EPIC-Oxford study, formulated the hypothesis of this study, and edited the manuscript. None of the authors had any conflicts of interest with regard to the organizations sponsoring the research.

REFERENCES

- Ballantyne CM. Low-density lipoproteins and risk for coronary artery disease. *Am J Cardiol* 1998;82:3Q-12Q.
- Clarke R, Lewington S, Youngman L, Sherliker P, Peto R, Collins R. Underestimation of the importance of blood pressure and cholesterol for coronary heart disease mortality in old age. *Eur Heart J* 2002;23:286-93.
- Lloyd-Jones DM, Larson MG, Beiser A, Levy D. Lifetime risk of developing coronary heart disease. *Lancet* 1999;353:89-92.
- Bittner V. Lipoprotein abnormalities related to women's health. *Am J Cardiol* 2002;90(suppl):77i-84i.
- Clarke R, Frost C, Collins R, Appleby P, Peto R. Dietary lipids and blood cholesterol: quantitative meta-analysis of metabolic ward studies. *BMJ* 1997;314:112-7.
- Anderson JW, Johnstone BM, Cook-Newell ME. Meta-analysis of the effects of soy protein intake on serum lipids. *N Engl J Med* 1995;333:276-82.
- Wangen KE, Duncan AM, Xu X, Kurzer MS. Soy isoflavones improve plasma lipids in normocholesterolemic and mildly hypercholesterolemic postmenopausal women. *Am J Clin Nutr* 2001;73:225-31.
- Sanders TAB, Dean TS, Grainger D, Miller GJ, Wiseman H. Moderate intakes of intact soy protein rich in isoflavones compared with ethanol-extracted soy protein increase HDL but do not influence transforming growth factor β_1 concentrations and hemostatic risk factors for coronary heart disease in healthy subjects. *Am J Clin Nutr* 2002;76:373-7.
- Merz-Demlow BE, Duncan AM, Wangen K, E et al. Soy isoflavones improve plasma lipids in normocholesterolemic, premenopausal women. *Am J Clin Nutr* 2000;71:1462-9.
- Crouse JR III, Morgan T, Terry JG, Elis J, Vitolins M, Burke GL. A randomized trial comparing the effect of casein with that of soy protein containing varying amounts of isoflavones on plasma concentrations of lipids and lipoproteins. *Arch Intern Med* 1999;159:2070-6.
- Uesugi T, Fukui Y, Yamori Y. Beneficial effects of soybean isoflavone supplementation on bone metabolism and serum lipids in postmenopausal Japanese women: a four-week study. *J Am Coll Nutr* 2002;21:97-102.
- Gardner CD, Newell KA, Cherin R, Haskell WL. The effect of soy protein with or without isoflavones relative to milk protein on plasma lipids in hypercholesterolemic postmenopausal women. *Am J Clin Nutr* 2001;73:728-35.
- Lichtenstein AH, Jalbert SM, Adlercreutz H, et al. Lipoprotein response to diets high in soy or animal protein with and without isoflavones in moderately hypercholesterolemic subjects. *Arterioscler Thromb Vasc Biol* 2002;22:1852-8.
- Jenkins DJA, Kendall CWC, Jackson C-JC, et al. Effects of high- and low-isoflavone soyfoods on blood lipids, oxidized LDL, homocysteine, and blood pressure in hyperlipidemic men and women. *Am J Clin Nutr* 2002;76:365-72.
- Nestel PJ, Yamashita T, Sasahara T, et al. Soy isoflavones improve systemic arterial compliance but not plasma lipids in menopausal and perimenopausal women. *Arterioscler Thromb Vasc Biol* 1997;17:3392-8.
- Dewell A, Hollenbeck CB, Bruce B. The effects of soy-derived phytoestrogens on serum lipids and lipoproteins in moderately hypercholesterolemic postmenopausal women. *J Clin Endocrinol Metab* 2002;87:118-21.

17. Hodgson JM, Puddey IB, Beilin LJ, Mori TA, Croft KD. Supplementation with isoflavonoid phytoestrogens does not alter serum lipid concentrations: a randomized controlled trial in humans. *J Nutr* 1998;128:728–32.
18. Simons LA, von Konigsmark M, Simons J, Celermajer DS. Phytoestrogens do not influence lipoprotein levels or endothelial function in healthy, postmenopausal women. *Am J Cardiol* 2000;85:1297–301.
19. Weggemans RM, Trautwein EA. Relation between soy-associated isoflavones and LDL and HDL cholesterol concentrations in humans: a meta-analysis. *Eur J Clin Nutr* 2003;57:940–6.
20. Davey GK, Spencer EA, Appleby PN, Allen NE, Knox KH, Key TJ. EPIC-Oxford: lifestyle characteristics and nutrient intakes in a cohort of 33 883 meat-eaters and 31 546 non meat-eaters in the UK. *Public Health Nutr* 2003;6:259–69.
21. Verkasalo PK, Appleby PN, Davey GK, Key TJ. Soy milk intake and plasma sex hormones: a cross-sectional study in pre- and postmenopausal women (EPIC-Oxford). *Nutr Cancer* 2001;40:79–86.
22. Holland B, Unwin ID, Buss DH. Milk products and eggs. Fourth supplement to McCance and Widdowson's *The composition of foods*, 4th ed. Cambridge, United Kingdom: Royal Society of Chemistry, 1989.
23. Holland B, Unwin ID, Buss DH. Vegetables, herbs and spices. Fifth supplement to McCance and Widdowson's *The composition of foods*, 4th ed. Cambridge, United Kingdom: Royal Society of Chemistry, 1991.
24. Holland B, Welch AA, Buss DH. Vegetable dishes. Second supplement to McCance and Widdowson's *The composition of foods*, 5th ed. Cambridge, United Kingdom: Royal Society of Chemistry, 1992.
25. US Department of Agriculture-Iowa State University. Database on isoflavone content of foods. 1999. Internet: <http://warp.nal.usda.gov/fnic/foodcomp/Data/isoflav/isoflav.html> (accessed 1 Apr 2004).
26. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem* 1972;18:499–502.
27. Verkasalo PK, Appleby PN, Allen NE, Davey GK, Adlercreutz H, Key TJ. Soy intake and plasma concentrations of daidzein and genistein: validity of dietary assessment among eighty British women (Oxford arm of the European Prospective Investigation into Cancer and Nutrition). *Br J Nutr* 2001;86:415–21.
28. Bingham SA, Gill C, Welch A, et al. Validation of dietary assessment methods in the UK arm of EPIC using weighed records, and 24-hour urinary nitrogen and potassium and serum vitamin C and carotenoids as biomarkers. *Int J Epidemiol* 1997;26(suppl):S137–51.
29. Anderson JW, Smith BM, Washnock CS. Cardiovascular and renal benefits of dry bean and soybean intake. *Am J Clin Nutr* 1999;70(suppl):464S–74S.
30. Nagata C, Takatsuka N, Kurisu Y, Shimizu H. Decreased serum total cholesterol concentration is associated with high intake of soy products in Japanese men and women. *J Nutr* 1998;128:209–13.
31. de Kleijn MJJ, van der Schouw YT, Wilson PWF, Grobbee DE, Jacques PF. Dietary intake of phytoestrogens is associated with a favourable metabolic cardiovascular risk profile in postmenopausal U.S. women: the Framingham Study. *J Nutr* 2002;132:276–82.
32. Goodman-Gruen D, Kritiz-Silverstein D. Usual dietary isoflavone intake is associated with cardiovascular disease risk factors in postmenopausal women. *J Nutr* 2001;131:1202–6.
33. Posner BM, Cupples LA, Miller DR, et al. Diet, menopause, and serum cholesterol levels in women: the Framingham Study. *Am Heart J* 1993;125:483–9.
34. Zhang X, Shu XO, Gao Y-T, et al. Soy food consumption is associated with lower risk of coronary heart disease in Chinese women. *J Nutr* 2003;133:2874–8.
35. Nagata C, Takatsuka N, Shimizu H. Soy and fish oil intake and mortality in a Japanese community. *Am J Epidemiol* 2002;156:824–31.
36. Food and Drug Administration. Food labeling: health claims; soy protein and coronary heart disease. Final Rule. *Fed Regist* 1999;64:57700–33.

