Food Frequency Questionnaires and Overnight Urines Are Valid Indicators of Daidzein and Genistein Intake in U.S. Women Relative to Multiple 24-h Urine Samples

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Data regarding convenient, valid methods for measuring U.S. isoflavone intake are limited. We evaluated a soy food questionnaire (SFQ), the Willett food frequency questionnaire (FFQ), and overnight urine samples relative to excretion in 24-h urine samples. We also described intake among women in a high-risk program for breast or ovarian cancer. Between April 2002 and June 2003, 451 women aged 30 to 50 yr with a family history of breast or ovarian cancer completed the SFQ and FFQ. Of them, 27 provided four 24-h and overnight urine specimens. In these women, 24-h sample measures were correlated with SFQ estimates of daidzein (Spearman $r = .48$) and genistein ($r = .54$) intake, moderately correlated with the Willett FFQ (daidzein $r = .38$, genistein $r = .33$), and strongly correlated with overnight urine excretion (daidzein $r = .84$, genistein $r = 0.93$). Among all 451 SFQ respondents, mean (median) daidzein and genistein intakes were 2.8 (0.24) and 3.9 (0.30) mg/day. Primary sources of both were soymilk, soy nuts, and tofu. We conclude that targeted soy food questionnaires, comprehensive FFQs, and multiple overnight urines are all reasonable options for assessing isoflavone intake in epidemiologic studies.

INTRODUCTION

Ecologic (1), epidemiologic (2), and experimental (3) research has suggested that soy or isoflavone intake can reduce risk of breast cancer, but whether current levels of isoflavone intake in the usual U.S. diet are sufficiently high to affect breast cancer risk remains unclear. The marketing of soy as a health food and the greater popularity of nonmeat protein alternatives have contributed to a substantial rise in the availability and purchase of soy-containing products (4–6). Although statistics on isoflavone intake are not available, sales of soy-based products increased 20% per year since 1995 compared with <10% before 1995 (7). The increase may be especially pronounced in health-conscious individuals, such as women with a family history of breast cancer, who are likely more motivated to modify their lifestyle to reduce their breast cancer risk (8).

A primary obstacle to describing intake and health effects of isoflavones consumed in a population is the lack of a convenient

and validated method for measuring intake. Only two studies, to our knowledge, have evaluated the relative validity of such a dietary instrument against isoflavone excretion in multiple 24-h urine samples (9,10). Several studies have used spot or overnight urine excretion as alternatives because of their ease of implementation relative to 24-h samples, but one study that compared overnight urine excretion to 24-h samples found only moderate correlations between the two measures (9).

The objectives of this study were to evaluate the validity and reproducibility of a soy food questionnaire (SFQ) developed for non-Asian women; to evaluate two other methods—the Harvard Diet Assessment Form (DAF) (11) and multiple overnight urine samples—for their adequacy in ranking women relative to 24-h urine samples; and to use the SFQ to describe levels of intake and main food sources of isoflavones in a sample of women enrolled in a program for women at potentially high genetic risk for breast or ovarian cancer.

MATERIALS AND METHODS

Study Sample

Subjects in this study were drawn from the Family Risk Assessment Program (FRAP) at the Fox Chase Cancer Center (FCCC) in Philadelphia. FRAP was initiated in 1991 to offer education and preventive interventions to women with at least one first- or second-degree relative with breast or ovarian cancer. Recruitment strategies include referrals from breast or ovarian cancer patients at FCCC, radio and newspaper advertisements, and physician and self-referrals. On enrollment, women complete a health history questionnaire that elicited information on sociodemographic factors, reproductive factors, and family and medical history.

Between April 2002 and June 2003, an SFQ and the 126-item DAF (11) were sent to each of 893 FRAP subjects between the ages of 30 and 50 yr; 3 women had passed away, and 7 women were determined to be ineligible (due to having participated in a related study), leaving 883 potential respondents. Of 883 potentially eligible respondents, 371 did not return packets, 44 could not be contacted by mail or telephone, and 17 indicated that they were not interested in participating, leaving a sample of 451 respondents. Of these, a subset of 27 women agreed to provide urine specimens for comparison with questionnaire estimates.

The protocol for this research was reviewed and approved by the Fox Chase Cancer Center Institutional Review Board.

Assessment of Isoflavone Intake

We developed an SFQ and an accompanying database of isoflavone content values to estimate frequency of intake of soy-containing foods and level of intake of daidzein and genistein. A detailed description of the SFQ has been provided previously (12). Briefly, the SFQ followed the format of the DAF. The list of items in the SFQ and its accompanying database of isoflavone values were developed from tables and databases representing all currently available information on the isoflavone content of over 100 food items (13–18). These data were supplemented with information obtained from soy food manufacturers and with detailed soy intake information from pilot testing in 12 women who completed a draft version of the soy food questionnaire and were then interviewed for qualitative feedback on the questionnaire and on their soy food eating habits in general. Their responses led to deletion of infrequently eaten items from the food list (e.g., soy pizza) and to refinement of foods included as the basis for the isoflavone database.

For each food item, subjects indicated the average frequency that they consumed a specified portion size of the food over the past year, with 9 frequency options ranging from "never, or less than once per month" to "6+ per day." For soy protein bars and powders, we estimated mean frequency of intake based on the respondent's frequency of consumption of any protein bars/powders and how often the protein bars/powders were soy protein based. We then estimated frequency of intake soy protein bars/powders as 0, 0.33, 0.67, or 1.0 times the frequency of all protein bars/powders if women reported that the protein bars/powders they consumed were, respectively, "never or almost never," "sometimes," "often," or "always or almost always" soy protein based. When women reported that they did not know, we assumed that the product did not contain soy. The questionnaire also included an open-ended section asking about intake of soy supplements and of other soy-based foods eaten at least once a month. To estimate daidzein and genistein intake from the SFQ, intake of specific isoflavones was estimated by multiplying frequency of intake of each food by the isoflavone content for the food's portion size.

We also estimated daidzein and genistein intake based on responses to the DAF using a previously described method (19). In this method, isoflavone values were assigned to each food item in the DAF using values identified in the literature and unpublished data from experts in the field. The phytoestrogen content of each food item was assigned a score (0, 0.0005, 0.005, 0.05, 0.5, 5, or 50 mg/100 g wet weight of food) to avoid implying a degree of accuracy for which current data on isoflavone content are too limited. The score for each food item was then multiplied by the serving size and frequency of consumption of the food and then summed across foods to estimate total intake.

Assessment of Urinary Phytoestrogens

From 27 women who agreed to provide urine samples, four 24-h urine specimens were collected for four 24-h periods, each beginning with the sample following the first void of the selected day, over the period of 1 mo. Two samples were requested during each half of each woman's menstrual cycle. Subjects received 4-l containers with 4 g of ascorbic acid to prevent microbial contamination and oxidative degradation. They were asked to use a separate container for the overnight urine sample, or the

first void of the next day, and to store the overnight and rest-of-day sample containers in their refrigerators until pick-up the next morning.

On transport to the FCCC, total 24-h urine volume was recorded, and sodium azide was added to achieve a 0.1% (wt/vol) concentration. After extracting separate 25-ml aliquots of overnight samples, remaining overnight samples were combined with rest-of-day samples in correct proportions to create pooled 24-h samples, which were also put into 25-ml aliquots. All samples were stored at –20°C until shipment to the laboratory of Dr. Mindy Kurzer at the University of Minnesota for phytoestrogen analyses or to Quest Diagnostics for analysis of urinary creatinine using a kinetic colorimetric assay [interassay coefficients of variation (CV) of 2–3%].

Procedures for analyzing urinary specimens for phytoestrogens have been described (20). Briefly, immediately before analysis, urine aliquots were thawed. Each of the 4 overnight samples were combined to create a pooled overnight urine sample. The four 24-h urine aliquots were similarly pooled. Fifteen-ml aliquots from each pooled overnight, and 24-hour urine samples were extracted.

Samples were analyzed for phytoestrogens, including the 3 major soy isoflavones (daidzein, genistein, glycitein) and 2 metabolites of daidzein [O-desmethylangolensin (ODMA) and equol], using an ion-exchange chromatography and capillary gas chromatography-mass spectrometry method (21) in which phytoestrogens were first extracted on Bond Elut C_{18} columns. Deuterated phytoestrogen and estrogen internal standards were added to each sample. The aliquots were then hydrolyzed with *Helix pomatia* enzyme extract. The isoflavonoid fraction was separated from the rest of the compounds on the acetate form of QAE-Sephadex columns. Trimethylsilyl derivatives of the samples and standards were analyzed by a Hewlett Packard 5890 and 5971A quadrupole gas chromatography-mass spectrometry instrument operated with a Unix 59940A ChemStation and a HP 7673 autosampler (Agilent Technologies, Santa Clara, CA) in the selective ion-monitoring mode. Intraassay CVs ranged from 4.0–10.8%, and interassay CVs ranged from 9.7–23.5% for analyses of the 5 phytoestrogens examined here. All samples from each subject were analyzed in duplicate in the same batch.

Statistical Analyses

We used Spearman correlations to compare isoflavone estimates from the SFQ, estimates from the DAF, and urinary isoflavone excretion in overnight urine samples with 24-h urine samples (in nmol/mg creatinine).

In addition to estimating total daily daidzein and genistein intake for each respondent, we calculated total daidzein and genistein intake over the entire sample as the sum of daidzein and genistein intake over all respondents. This sum was then used to calculate the contribution of each individual soy food from the SFQ to total daidzein or genistein intake—specifically, by calculating the amount from the particular food summed over all respondents then dividing by total intake for the entire sample.

All analyses were conducted using Statistical Analysis Software (SAS) release 9.1 (SAS Institute Inc., Cary, NC).

RESULTS

Characteristics of the sample that responded to the SFQ ($N = 451$) and of the subset that provided urine specimens ($N = 27$) are shown in Table 1. Mean (SD) age of the full sample of respondents ($N = 451$) was 41.9 yr (5.3), most (96%) women were White, and most (66%) had attained at least a college degree. The subsample ($N = 27$) was similar to the full sample with respect to sociodemographic characteristics as well as intake of energy, fat, and fiber. The SFQ and DAF gave different pictures of daidzein and genistein intake in the full and subsamples, however. Whereas the SFQ estimated higher median intake of the two isoflavones in the full sample compared with the subsample, the DAF showed similar estimates of median intake between the two samples. With respect to urinary measures, daidzein was the most abundant isoflavone detected in both 24-h and overnight samples, followed by genistein.

Daidzein and genistein intake estimated from the SFQ was correlated with their excretion in 24-h urine specimens (Table 2). The correlation for daidzein was similar when daidzein metabolites (ODMA and equol) were included (Spearman $r = .49$). To exclude the influence of extreme observations on our correlations, we excluded four subjects with 0 isoflavone intake based on the SFQ and 1 subject with isoflavone intake >100 mg/day. Excluding these subjects substantially increased Spearman correlations for both daidzein ($r = .63$) and genistein ($r = .63$). In an examination of individual items in the SFQ, the three that were most highly correlated with daidzein excretion were soymilk ($r = .51$), vegetable burgers ($r = .50$), and soy nuts ($r = .33$), whereas the three most highly correlated with genistein excretion were soymilk ($r = .50$), vegetable burgers ($r = .48$), and tofu ($r = .38$). Correlations for estimates based only on two items, soymilk and vegetable burgers, were .61 for daidzein and .60 for genistein. Excluding the four subjects with 0 isoflavone intake and the one subject with isoflavone intake >100 mg/day did not change the correlations nor did adding in estimates from other individual food items meaningfully improve these correlations (results not shown).

Correlations for daidzein and genistein intake estimated from the DAF were weaker but still moderate when compared with 24-h urine sample measures (Table 2). When daidzein metabolites were also included, the correlation for daidzein improved considerably ($r = .54$). Isoflavones measured in overnight urines were strongly correlated with their corresponding measures in 24-h samples (Table 2). The correlation between overnight and 24-h samples for all 5 isoflavones summed together was .91.

We estimated isoflavone intake among the full sample of respondents to the questionnaire ($N = 451$). Mean (SD) daily intakes of daidzein and genistein estimated from the SFQ were

TABLE 1
Characteristics of All Respondents to Soy Food Questionnaire and Subset of Participants Providing Urine Specimens[a]

	Main Study Sample ($N = 451$)	Subsample ($N = 27$)
Mean (SD) age (yr)	41.9 (5.3)	41.3 (4.5)
White (%)	96	96
Highest grade completed (%)[b]		
<College degree	33	22
College degree	40	56
>College degree	27	22
Mean (SD) BMI (kg/m^2)	—	25.4 (6.5)
Mean (SD) daily intake		
Energy (kcal)	7,622 (2805)	7,803 (2562)
Energy from fat (%)	32 (7)	32 (5)
Fiber (mg)	19 (10)	19 (8)

	Main Study Sample		Subsample	
Isoflavone Intake (mg/day)	Mean (SD)	Median (Interquartile Range)	Mean (SD)	Median (Interquartile Range)
Daidzein				
SFQ	2.8 (7.4)	0.237 (0–1.485)	2.9 (9.4)	0.015 (0.007–1.310)
DAF	2.3 (8.7)	0.029 (0.020–0.058)	2.1 (4.6)	0.032 (0.023–3.331)
Genistein				
SFQ	3.9 (10.4)	0.302 (0–1.972)	3.9 (12.2)	0.013 (0.006–2.000)
DAF	2.4 (8.8)	0.097 (0.050–0.325)	2.2 (4.6)	0.129 (0.053–3.438)
Urinary excretion (ng/mg creatinine)				
24-h urine				
Daidzein	—	—	313.4 (618.9)	110.3 (52.2–428.4)
Genistein	—	—	136.4 (161.2)	67.4 (25.6–201.4)
Glycitein	—	—	42.6 (84.2)	16.2 (3.6–39.9)
ODMA	—	—	88.9 (208.5)	18.9 (6.5–44.5)
Equol	—	—	122.9 (425.2)	12.8 (2.3–29.1)
Overnight urine				
Daidzein	—	—	419.7 (885.6)	135.0 (87.8–397.4)
Genistein	—	—	250.5 (476.3)	77.2 (33.5–264.5)
Glycitein	—	—	58.1 (139.3)	11.1 (3.3–44.6)
ODMA	—	—	93.4 (232.0)	15.5 (8.7–70.6)
Equol	—	—	84.3 (274.1)	10.0 (7.2–20.9)

[a]All respondents to soy food questionnaire, $N = 451$; participants providing urine specimens, $N = 27$. Abbreviations are as follows: BMI, body mass index; SFQ, soy food questionnaire; DAF, Harvard Diet Assessment Form; ODMA, O-desmethylangolensin.
[b]Due to missing data, education level is described for $N = 446$ in the full study sample.

2.8 (7.4; median = 0.24) and 3.9 (10.4; median = 0.30) mg/day, respectively, higher than estimates based on the DAF (for daidzein, mean = 2.3, SD = 8.7, median 0.03; for genistein, mean = 2.4, SD = 8.8, median 0.10). The 10 top sources of the two isoflavones (genistein + daidzein) based on the SFQ are shown in Table 3. Together, these sources accounted for 87% of total daidzein and genistein intake in our sample. Although soymilk was the most important contributor to total isoflavone intake in the sample, more women (30%) consumed vegetable burgers than any other soy food or soy-containing food. Among women reporting consumption of any given food, soy milk and soy cereal were the most frequently consumed.

DISCUSSION

Our findings extend the field of isoflavone intake assessment in several ways. First, we found that our 26-item SFQ performed well in ranking women according to their daidzein and genistein intake compared with multiple 24-h urine specimens, although correlations were even higher for estimates based on

TABLE 2
Spearman Correlations With Isoflavone Levels (ng/mg Creatinine) in Multiple 24-h Urine Samples[a]

	SFQ	DAF	Multiple Overnight Urine Samples
Daidzein	.48	.38	.84
Genistein	.54	.33	.93
Glycitein	—	—	.76
ODMA	—	—	.84
Equol	—	—	.61

[a]$N = 27$. Abbreviations are as follows: SFQ, soy food questionnaire; DAF, Harvard Diet Assessment Form; ODMA, O-desmethylangolensin.

only 2 of the 26 items (soymilk and vegetable burgers). Thus, our work offers ideas for assessing isoflavone intake using a potentially reducible soy questionnaire module appropriate for a non-Asian, Eastern U.S. population. Second, our analysis is among the first to demonstrate that a comprehensive FFQ yields reasonable correlations with excretion in 24-h urine samples. Our analysis is also among the first to demonstrate that multiple overnight urine samples are strongly correlated with 24-h urine samples—clearly more strongly correlated than were any of the questionnaire-based estimates. Many studies have used overnight urines as a referent to evaluate questionnaire-based isoflavone estimates, but only one study, to our knowledge, evaluated overnight urine samples by comparing them against a more rigorous referent, multiple 24-h urine samples, and that study showed substantially more modest correlations than did our data (9). Finally, our study is the first to describe isoflavone intake in a sample of women at potentially high genetic risk for breast or ovarian cancer. In this sample, we found generally low levels of isoflavone intake. Top contributors to total isoflavone intake were soymilk, soy nuts, and tofu, but vegetable burgers were consumed by more women in our sample than any other soy food.

TABLE 3
Ten Top Sources of Dietary Daidzein and Genistein Among Respondents to the SFQ[a]

	Total genistein + Daidzein Intake in Sample (%)[b]	Respondents Consuming at Least Once per Month (%)	Mean Monthly Frequency of Consumption Among Consumers of the Food
Soy milk	27.5	14	16
Soy nuts	16.7	13	8
Tofu	10.4	17	5
Soy bars	10.1	10	7
Soy powders	8.2	6	9
Vegetable burgers	4.3	30	4
Green soybeans	2.8	12	3
Soy yogurt	2.6	2	8
Soybeans	2.2	4	2
Soy cereal	2.0	3	12
Total	86.7		

[a]$N = 451$. Abbreviation is as follows: SFQ, soy food questionnaire.
[b]Calculated as amount of daidzein + genistein from the individual food summed over all respondents divided by total daidzein and genistein intake over entire sample.

Our SFQ compares favorably with other such questionnaires developed for non-Asian samples. Correlations between questionnaire estimates and either urine (22) or plasma (23,24) samples were .3 for a 6-item module (22), .4 for a 20-item module (23), and .5 for a 40-item module (24), all developed for samples in Washington state. The correlation with an overnight urine sample for an isoflavone intake estimate based on 12 soy-containing foods was .3 for annual and .6 for recent soy intake in a multiethnic sample in Hawaii (25); but in a subsequent analysis, the tetrachoric correlation for the same questionnaire was .88 including both control and intervention subjects in a soy intervention study (26). Estimates based on 6 to 8 soy-containing items within a FFQ were moderately correlated ($r = .2$–$.4$) with either urine or serum measures in studies conducted in Japan (27) and China (28,29).

We found a correlation of $r = .6$ when we based isoflavone estimates on only two items (soymilk and vegetable burgers) compared with a correlation of $\approx .5$ based on all the soy foods in the questionnaire. Because we would expect that including more soy foods would improve estimates and correlations, this finding indicates substantial error in estimating isoflavone intake from other soy food items. The source of error is not clear and could include inaccurate recall of dietary intake, inaccurate isoflavone content values, or differences in the bioavailability of isoflavones from different food sources (30). Lampe et al. (10) observed correlations of only $\approx .2$ between daidzein and genistein excretion in multiple 24-h urine samples and soy intake estimated from a single FFQ item (tofu or soybeans). Other researchers, however, have found that 2 to 3 soy food items are sufficient to rank individuals based on their isoflavone intake. Verkasalo et al. (31) found a strong correlation ($r = .7$) with plasma concentrations for daidzein and genistein estimates based on two items (tofu, soy foods) in a sample of women that was selected to represent a wide range of soy food intake. In work by Frankenfeld et al. (23,24), correlations with plasma measures for daidzein and genistein estimates based on two items (tofu and soymilk) were only slightly lower than those observed for 20- and 40-item SFQs. Similarly, Yamamoto et al. (27) found that 3 soy food items were sufficient to yield correlations with a 24-h urine measure that were comparable to those based on the original 8 items.

Together, these findings suggest that a small number of soy-containing items is sufficient to explain variability in isoflavone intake and that an extensive module to assess soy food intake is probably not necessary. Nevertheless, selecting the appropriate items for a given population requires continued monitoring of soy food trends, especially given rapid changes in eating habits, soy product availability, and isoflavone content. In the United States, for example, whereas soymilk and tofu have remained relatively constant sources of isoflavones in the American diet since the mid-1990s (15,24,32), sales of tofu have recently leveled, and soy snacks are emerging as a popular, new category of soy foods (33,34). Indeed, soy nuts, although they have not been reported as a significant food item in earlier work (15,24,32), were a primary isoflavone source in our sample, and many subjects also reported consumption of soy chips (data not shown). Selecting the soy food items to include in a module may also require consideration of some statistical issues. In our sample, for example, vegetable burgers were the second most highly correlated but contributed only 4.3% to total isoflavone intake, suggesting that it was able to capture intraindividual variability in isoflavone intake despite its relatively moderate contribution to total isoflavone intake.

The correlations that we observed for the DAF were comparable to those obtained in previous work using other comprehensive FFQs. This was despite the use of an alternative method of assigning phytoestrogen values to FFQ food items that assigned categories instead of exact phytoestrogen content values (19). In a study including 51 Japanese American women, the correlations for daidzein and genistein intake estimated from a modified Block FFQ were .49 and .30, respectively, using multiple, 24-h urine samples as the referent (9). In another study of 58 South Asian women, correlations for intake estimated from a 207-item FFQ were .32 and .21 for daidzein and genistein, respectively, with multiple plasma samples as the referent (35). These findings suggest that a comprehensive FFQ can provide a reasonable ranking of individuals based on their isoflavone intake even when the FFQ does not specifically target soy foods, indicating the importance of nonsoy sources of isoflavones (19,32). Among postmenopausal women in the Framingham Offspring Study, for example, soy (tofu and soybeans) contributed only 3.3% to total isoflavone intake, whereas beans and peas, tea, coffee, and nuts were the main contributors, together accounting for >55% of total intake (19). How well an open-ended section in such FFQs can improve correlations is unclear. Ideally, at least one or two soy food items should be added to improve estimates.

Only 2 other studies, to our knowledge, have compared FFQ estimates of isoflavone intake against excretion in multiple, 24-h urine specimens (9,10). Others have used measures in a single, 24-h sample (27), in overnight or spot urine samples (22,25,28), or in serum or plasma (23,24,31,35). How well such measures reflect intake is unclear given that daidzein and genistein have a half-life of ≈8 h and are mostly excreted within 12 h of ingestion (36). In our sample, isoflavone measures in overnight urine measures were generally very highly correlated with 24-h urine measures—more strongly correlated than our questionnaire-based estimates and also more strongly correlated than was reported in previous work (9). Although our study procedures did not permit an assessment of the adequacy of 1 vs. multiple overnight urines, a previous study found no significant difference in isoflavone excretion comparing 2 separate overnight samples in a sample of women in Washington State (22). In another study, the analysis of first morning urine collected once, combined from 3 days a week and combined from 3 days a week over 1 mo, all showed excellent correlations, indicating that urine from a single overnight collection is as good as one collected over 3 days per week or over 1 entire mo (37). Thus, for ranking of isoflavone intake and excretion, overnight urine samples represent a good alternative to 24-h urine samples. Collection of 24-h urine samples poses a burden to study subjects as well as study organizers and can result in bias because of the high potential for both nonparticipation and noncompliance among subjects. In general, when working with a large, population-based sample, using the SFQ or a similar questionnaire may be more feasible than obtaining urine-based assessments of soy intake. However, when obtaining overnight samples is feasible, or when working with a low-literacy population, overnight urines may be the preferable method.

Our results provide the first description, to our knowledge, of isoflavone intake in women at potentially high genetic risk for breast or ovarian cancer. When we used the SFQ to describe intake in this sample, we found estimates of intake that were generally lower than those for samples in California (15), Washington (23,24), and Hawaii (25). Possibly, these women have controlled their soy intake as a result of concerns about the estrogenic effects of isoflavones. In a previous analysis, however, only 7% of nonconsumers wrote that they avoided soy foods for this reason (12). In our previous analysis, we found that not knowing how to prepare soy foods and disliking the taste of soy foods were the primary reasons for their nonconsumption (12). These reasons are the most likely explanations for our subjects' generally low level of isoflavone intake and for their more frequent consumption of vegetable burgers in favor of richer but less familiar isoflavone sources such as tofu.

The scope of our work directly addressed only the validity of questionnaires and overnight urine samples for assessing isoflavone intake in epidemiologic studies. Budgetary constraints necessitated the pooling of the four 24-h and overnight urine samples for laboratory analyses, thus limiting our ability to evaluate interindividual variability in the isoflavone content of urine samples or the validity of analyzing single, overnight urine samples as a more convenient measure of isoflavone intake. Evaluating measures to estimate absolute level of intake in individuals as opposed to relative intake within a population was also beyond the scope of our analysis. These merit investigation in future studies.

Findings are subject to bias if subjects were systematically different from nonsubjects. In a previous analysis (12) we found that respondents to the SFQ were slightly older than

nonrespondents (mean age = 42 vs. 41 yr) and more likely to be college educated (66% vs. 57%), but they were similar with respect to other demographic characteristics and health behaviors. We observed no significant differences in demographic characteristics or health behaviors, including isoflavone intake, between SFQ respondents ($N = 451$) and subsample subjects ($N = 27$).

Also worth noting is that our sample consisted of subjects in a program for women with a family history of breast or ovarian cancer; thus, our estimates of soy and isoflavone intake apply to more health-conscious, potentially high-risk populations. We have no reason to expect, however, that the correlations we observed between questionnaire-based estimates and isoflavones excreted in the urine would differ from average-risk women. Thus, we believe that our conclusions regarding measurement of soy and isoflavone intake are applicable to a broader population.

In conclusion, we found that a 26-item SFQ performed well in ranking women based on their daidzein and genistein intake. As few as two items, however, perform as well or better, but selection of the items requires consideration of trends in soy food availability and consumption and validation in an appropriate sample. We also found that a comprehensive FFQ not specifically designed to assess isoflavone intake also performed reasonably well and that multiple overnight urines were strongly correlated with 24-h urine measures. Our work provides several options for assessing isoflavone intake in epidemiologic studies.

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