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Relative Validity and Reproducibility of a Semi-quantitative Food Frequency Questionnaire among Urban Iranians

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ABSTRACT

The contribution of dietary factors to the development and prevention of non-communicable diseases is being increasingly recognized, and worldwide, the concern about chronic diseases is growing rapidly. There is a lack of a comprehensive food frequency questionnaire (FFQ) encompassing all kinds of foods for Iranians. This cross-sectional study aims to describe the relative validity and reproducibility of the FFQ used for assessing nutrient intakes of Iranian urban population. One hundred thirteen subjects aged 20–69 years from five major cities of Iran participated in this study. The dietary intake was assessed by a semi-quantitative FFQ involving 160 Iranian food items. The participants were asked to complete two FFQs (at the first and fourth months of the study) and three-day food records every month. Two blood and 24-h urine samples were collected at fifth and sixth months. The highest correlation coefficient in all FFQs was for fat ($r = 0.669$; $p < 0.001$) and the lowest for fiber ($r = 0.331$; $p = 0.001$). The Pearson correlation coefficients between nutrient intake estimated by the average of the two FFQs and the average of the three-day food records ranged from 0.03 to 0.27. The Pearson correlation coefficients between serum and urine biomarkers and nutrient intake estimated by the average of the two FFQs ranged from -0.34 to 0.47 . Bland and Altman analyses showed fairly good agreement between the average of the FFQs and three-day food records for energy, fat, and potassium intakes. The obtained results indicate a reasonable validity of the FFQ considering the energy, fat, and potassium intake evaluation and good reproducibility over a 6-month period.

Introduction

The contribution of dietary factors to the development and prevention of non-communicable diseases is being increasingly recognized [1]. The worldwide concern about chronic diseases such as cancer and cardiovascular disease is growing rapidly [2]. The

analysis of dietary patterns has been revealed as a possible approach to examining the diet-disease relations [3]. The main methodological problem in observational studies of these relationships is the absence of a valid method for measuring dietary variables [4]. Obtaining an accurate estimate of long-term habitual food intake remains the main

challenge in diet-disease research [5]. Owing to its ease of administration and low burden on the subject, the assessment of dietary intake in epidemiological studies is often accomplished by means of food frequency questionnaires (FFQs) [6]. The FFQ is one of the most commonly used methods in epidemiological studies to assess individual long-term dietary intakes of foods and nutrients [7]. The underlying principle of the FFQ approach is that an average long-term diet, with consumption patterns over weeks, months, or years, is theoretically a more relevant determinant of chronic disease than the dietary intake pattern on a few specific days. Thus, more crude information relating to an extended period may be more useful than the precise intake measurements obtained on one or a few days. FFQs must be validated compared with more comprehensive and precise methods of assessment, such as diet records [8]. Because short-term recalls and diet records are generally expensive and unrepresentative of usual intake and not good for assessment of past diet, FFQ has been the primary method of dietary assessment for most epidemiological studies. FFQ is easy to manage and relatively inexpensive to use in large populations [9]. Recognizing its inherent potential for capturing usual dietary patterns and its intrinsic vulnerability to be affected by error [5], it is crucial to estimate the validity and reliability of an FFQ. For the interpretation of findings and comparability between studies, validation and calibration of FFQs are necessary in nutritional epidemiology. Validation studies show the ability of the method for truly measuring what it was designed to measure, whereas calibration studies determine how one method of dietary assessment compares with a reference method [7]. Since previous FFQs that have been designed for Iranian population consisted of specific kind of foods and for a certain types of diseases, such as diabetes, cardiovascular diseases, and esophageal cancers, a FFQ consisting of all kind of foods (traditional, western, etc.) was necessary for the complete evaluation of food intake of Iranian individuals. The aim of this study was to describe the relative validity and reproducibility of a novel FFQ used for assessing the nutrient intakes of the Iranian

urban population.

Method

Subjects

This cross-sectional study was conducted over a 6-month period. Study participants were chosen from five major cities of Iran (Tehran, Tabriz, Mashhad, Isfahan, and Shiraz); these cities belong to five provinces which are located in east/north-east, central, south, west/north-west, and capital of Iran, covering different ethnicities. Fall and winter seasons were selected as the period of study during which some subjects resigned from the study group and couldn't be substituted. During the last two-month interval of the total study period, 40 subjects were selected for performing biochemical tests. Probability proportional to size (PPS) sampling was used in this study. Firstly, five states were selected based on the population of each state; this selection was performed based on cumulative frequency and determining the interval based on 250 persons. For determining the interval, the whole population was divided by 5, and based on this interval, the states were selected. In each state, one city was selected based on the population of all cities of that state. The method by which the cities were selected was also based on PPS sampling and the population of each city; sample selection in each city was done randomly. In each city, two random areas were chosen, and then, one person (aged 20–69 years) from each family was selected. The sample size was calculated based on the approach applied by an analogous study (9). Based on the correlation coefficient between the concentrations of biomarkers and nutrient intakes as estimated by the FFQ, confidence interval of 95%, and study power of 80%, a sample size of 42 subjects for each city was required, totaling 210 ($42 \times 5 = 210$) subjects for the current study. Iranian nationality and residence in urban areas were the inclusion criteria for this study. For reducing the effect of over- and under-reporting, subjects who had reported a total energy intake beyond the range of 800–4200 kcal on FFQs were excluded. We also excluded those who did not

complete three-day food records. An informed written consent form was obtained from each subject. The study was approved by the research ethical committee of Mashhad University of Medical Sciences. We assessed the following variables in the present study: dietary intake of energy, carbohydrate, fat, protein, vitamin A, vitamin E, folate, and potassium; serum levels of vitamin A, vitamin E, and folate; and urinary concentrations of protein and potassium.

Dietary assessment

Food frequency questionnaire

For designing the final FFQ and choosing the food items, a preliminary FFQ was designed. This preliminary FFQ included a brief set of instructions, a food list of 302 items, three portion size options, eight frequency options, and an open-ended question to list food items which were consumed but not listed in the FFQ. This preliminary FFQ was trialed in 1011 participants (a group of employees from governmental and non-governmental organizations and their families). Following this step, food items that had maximum consumption of 1.5% were omitted from the FFQ. Food items that had minimum consumption of 5% were selected for the final FFQ, and other food items were combined with these items, resulting in a final FFQ containing 160 food items. For the validation study, the dietary intake was assessed by a semi-quantitative FFQ which consisted of 160 Iranian food items chosen by the methods described above. Based on the common average portion sizes among the Iranian population, the average consumption rate of the FFQ food items was determined. For explaining the exact amount of average consumption for the participants, we chose the commonly used serving items (e.g., bowl for yogurts and Chips; glass for beverages and plate for rice). Because similar dishes have different sizes, for illustrating average use, a food photo album consisting of 10 photos related to average uses of foods was placed at the beginning of the FFQ. The participants were asked to report their frequency of consumption of a given serving of each food item during the previous month. The food frequency consumption of each item was evaluated using nine categories:

never or less than once a month, 1 to 3 times a month, once a week, 2 to 4 times a week, 5 to 6 times a week, once a day, 2 to 3 times a day, 4 to 5 times a day, and 6 times or more a day. The food portions were classified into three sizes: small (half of the determined average use or less), medium (equal to the determined average use), and large (one half of average use or more). The participants completed the same FFQ twice, at the first month (FFQ1) and the fourth month of the study (FFQ2).

Three-day food record

A three-day food record was used as a reference method for validating the FFQ. During the 6-month period of the study, the participants were asked to complete food records monthly for three consecutive days including one weekend day. For explaining the portion sizes, a completed 1-day diet record was given as a sample to the participants; the photo album at the beginning of the FFQ was also another guide for them.

Nutrient analysis

In order to analyze the completed FFQs and reduce mistakes in recording FFQ data, a specific multi-function software was developed. First, all pages of the FFQs were scanned (HP Scan jet N8420 scanner) and then imported into a specific multi-function software developed. The first part of the software read the selected choices on the scanned pages of the FFQ and delivered an output file in TXT format. This part of the software was programmed by Borland Delphi 7. The second part of the software analyzed the data resulting from the first part and delivered the following data in an SPSS file: 1) which food items have been eaten; 2) how many grams of each food item has been eaten; 3) amount of consumed energy, macronutrients, fiber, and selected micronutrients (vitamin A, vitamin E, folate, and potassium). This part of the software was programmed by Microsoft Visual Basic .Net 2008. The presence of different recipes for each of food item was a problem; so we chose the most popular cooking book in Iran for reference [10]. First, the exact amount of the traditional food ingredients were measured in grams. Then, 100 grams of all the

needed ingredients based on the recipe in the selected cooking book were cooked and weighed again; the weight difference ratio after cooking was calculated for all the ingredients. The amount of energy, macronutrients, fiber, and mentioned micronutrients present in 100 grams of each food item of the FFQ were also included in the database of the mentioned software. The amount of these factors in the cooked ingredients were extracted from food composition tables [11] and calculated for traditional foods that were prepared based on the selected cooking book recipes. For some traditional foods that probably had different recipes being followed by people from different cities, we cooked the food based on Rosa Montazami's cooking book [10] and then sent them to the reference laboratory of food industry in Mashhad, Iran (Testa Lab) to measure their content of energy, macronutrients, and fiber. The completed three-day food records were analyzed using Nutritrac software [12].

Biochemical measurements

Due to the high expenses of measuring serum levels of micronutrients, only 40 individuals were selected to participate in the biochemical tests. One laboratory in each of the five cities was chosen, and the selected participants were referred to these laboratories. During the fifth and sixth months of the study, venous blood samples (5 ml) were taken from the brachial vein of the participants (who were on fast for sampling purposes) between 07:00 am to 09:00 am, and then the acquired samples were injected into the vacutainer tubes. While the blood samples were being taken according to a standard protocol, the individuals were in a sitting position. The blood samples were centrifuged at 3000 revolutions per minute (rpm) for 10 minutes. Serum was extracted and frozen at -20°C . The 24-h urine samples were also collected during the fifth and sixth months of the project in 1.5-liter plastic containers that were prefilled with 10 grams boric acid. Consequently, we collected two blood samples and two 24-h urine samples from each person. Vitamin A, vitamin E, and folate were measured in blood samples. Urinary

concentrations of potassium and protein were measured by electrolyte analyzer machine using colorimetry method.

We determined the validity of our FFQ through two methods: biochemical validation and use of a reference dietary assessment tool. The three-day food records were chosen as the reference tool for assessment of validity because they were expected to have a high answer level and good quality of response and not to interfere much with the normal dietary habits of the subjects. We evaluated the performance of the FFQ by comparing intake of nutrients and selected food groups obtained from this instrument with those derived from the 3-day food record.

Statistical analysis

For all statistical analyses, we used the SPSS statistical software package version 11.5. Kolmogorov-Smirnov test was used to assess the normality of the distributions of dietary intake variables. Means and standard deviations were calculated for energy, macronutrients, fiber, and selected micronutrient intakes from both FFQ and three-day food records. To show differences between the two FFQs, the Wilcoxon test and paired sample t-test were performed. Pearson correlation coefficients were used to determine correlations between two variables that had a normal distribution, such as nutrients (protein, vitamin A [retinol]), vitamin E (α -tocopherol), folic acid, and potassium intake, and their concentrations in serum and urine samples, as well as between FFQs and 3-day food records. We used the Bland-Altman method for assessing the agreement between FFQ and three-day food records among a range of nutrient intakes. In the present study, the differences of the two methods were plotted against three-day food records instead of the averages of the two methods since the three-day food record is a reference method for dietary intake assessment [13]. We interpreted the Bland-Altman results based on the Tang definition [14]. Good agreement is when the difference between the two methods of measurement is almost equal to one standard

deviation (SD) of the average nutrient intake from the reference method. Fairly good and bad/poor agreements are shown when the differences between the two measurements are almost equal to two and three SD of the average nutrient intake from the reference method, respectively. The reproducibility of the FFQ was evaluated by comparing the results of FFQ1 and FFQ2 by calculating the within-group correlation coefficients between them. A p-value <0.05 was considered significant for the present study.

Results

Two hundred ten adults aged 20–69 years (35 ± 12 years) from both genders participated in the current study, but because of the poor cooperation from the participants, only 113 subjects remained until the end of the project. Flow diagram of the number of individuals at each stage of the study is shown in Fig. 1. Thirty-nine subjects (34.5%) were men and 74 (65.4%) were women (Table 1). The mean and SD of the nutrient intakes determined by FFQ1 and FFQ2 and the average values of the three-day food records are reported in Table 2. Table 3, shows the correlation coefficients of nutrient intake between the two FFQs. Generally, correlation coefficients in men were higher than that of in women. The highest correlation coefficient in all FFQs was for fat ($r=0.669$; $p<0.001$) and the lowest one was for fiber ($r=0.331$; $p=0.001$). The correlation coefficients of the average nutrient intakes determined by the two FFQs and the three-day food records are reported in Table 4. The correlation coefficient for potassium was the highest ($r = 0.277$; $p = 0.019$). The serum and urine concentrations of the selected biomarkers (the average of two measurements) and the correlation coefficients between these biomarkers and the nutrient intake estimated by the average of the two FFQs are presented in table 5. The highest correlation coefficient was observed for potassium. The Bland-Altman analysis showed that compared with the three-day food record, the FFQ overestimated intakes for energy by 451.9 kcal/day (95% confidence interval [CI], -2102.2 to 1198.5), fat by 29.4 g/day (95% CI, -103 to 44.2), and potassium by 926.7 mg/day (95% CI, -4081.0 to 2227.6) (Fig. 2). Fig. 2. Bland-Altman

plots illustrating the relationship between average intake of a) energy, b) fat, and c) potassium, estimated using food frequency questionnaire (FFQ) and three-day food record methods ($n = 71$). The X axis is the amount of intake measured by the reference method (three-day food record). The Y axis is the difference between the intake data (energy, fat, and potassium) measured by FFQ (average of FFQ1 and FFQ2) and three-day food record (average of the six three-day food records). The central solid horizontal line indicates the mean difference between the two methods, and the solid lines above and below indicate ± 1.96 SD.

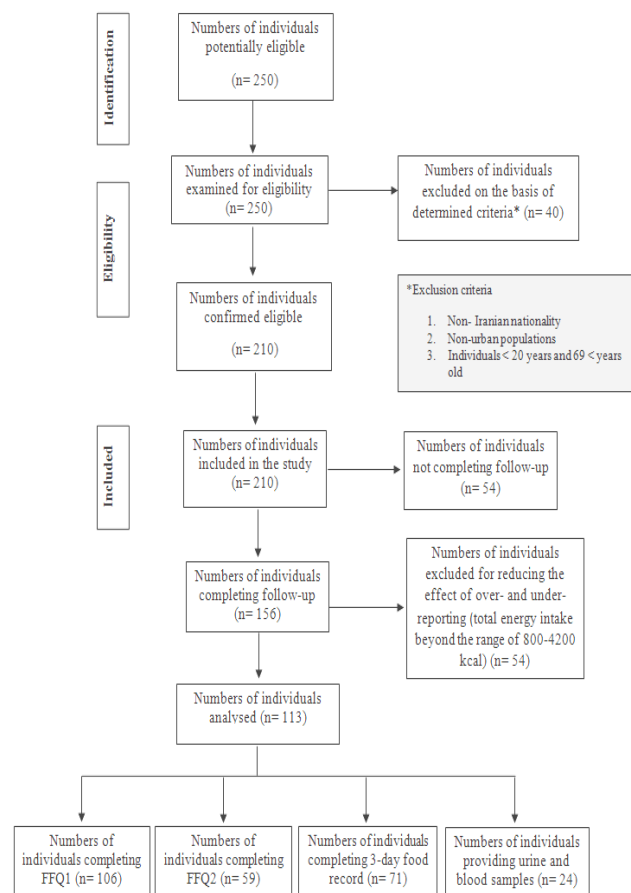


Figure 1. Flow Diagram of numbers of individuals at each stage of study

Table 1. Mean, standard deviation, minimum, and maximum of the age of the participants of the FFQ validation study (n = 113).

Variable	Sex	Number of subjects	Mean	Standard deviation	Minimum	Maximum
Age (years)	Man	39	38.75	13.99	20	69
	Woman	74	34.21	11.05	20	57
	Total	113	35.65	12.18	20	69

Table 2. Daily intake of energy and nutrients estimated by two FFQ and 3- day food record: FFQ validation study

Nutrient	FFQ1 (n=106)		FFQ2 (n=59)		3- day food record (n=71)	
	Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
Energy(Kcal)	2359.00	883.15	2304.60	823.60	2168.67	796.25
Carbohydrate (g)	291.44	111.95	280.45	100.44	294.81	130.56
Fat (g)	94.94	40.51	93.13	37.81	70.95	25.09
Protein (g)	86.39	32.29	87.90	37.69	86.62	28.158
Fiber (g)	21.18	12.00	21.33	10.27	24.38	25.70
Vitamin A (mcg)	865.67	480.47	981.08	528.57	1167.43	1116.00
Vitamin E (mg)	13.30	17.19	15.25	22.09	5.24	3.85
Folate (mcg)	454.73	315.55	478.02	273.10	482.05	252.82
Potassium (mg)	3660.80	1785.00	3645.40	1652.40	3161.33	1712.00

Table 3. Pearson correlation coefficients of nutrient intake estimated by two FFQs: FFQ validation study

Nutrient	Total (n=59)		Women (n=39)		Men (n=20)	
	Correlation coefficient (r)	P- value	Correlation coefficient (r)	P- value	Correlation coefficient (r)	P- value
Energy(Kcal)	0.575*	<0.001	0.515*	0.001	0.744*	<0.001
Carbohydrate (g)	0.456*	<0.001	0.409*	0.010	0.590	0.006
Fat (g)	0.669*	<0.001	0.600*	<0.001	0.863*	<0.001
Protein (g)	0.509*	<0.001	0.406**	0.010	0.727*	<0.001
Fiber (g)	0.357*	0.006	0.286	0.077	0.614*	0.004
Vitamin A (mcg)	0.409*	0.001	0.287	0.077	0.661*	0.002
Vitamin E (mg)	0.092	0.486	-0.002	0.989	0.759*	<0.001
Folate (mcg)	0.156	0.238	0.143	0.386	0.157	0.509
Potassium (mg)	0.456*	<0.001	0.355**	0.026	0.777*	<0.001

*Correlation is significant at the 0.01 level

** Correlation is significant at the 0.05 level

Table 4. Pearson correlation coefficients of nutrient intake estimated by the average of 3-day food records and the average of two FFQs (n=71): FFQ validation study

Nutrient	Correlation coefficient (r)	P- value	Nutrient	Correlation coefficient (r)	P- value
Energy(Kcal)	0.268	0.024	Vitamin A (mcg)	0.134	0.266
Carbohydrate (g)	0.206	0.084	Vitamin E (mg)	0.058	0.630
Fat (g)	0.247	0.038	Folate (mcg)	0.076	0.529
Protein (g)	0.207	0.084	Potassium (mg)	0.277	0.019
Fiber (g)	0.032	0.788			

Table 5. Mean and standard deviation of serum or urine concentrations of selected biomarkers and Pearson correlation coefficients between serum and urine biomarkers and nutrient intake estimated by the average of two FFQs (n=24): FFQ validation study

Nutrient	Mean	Standard deviation	Correlation coefficient (r)	P- value
Protein(mg/24 hr)	70.04	81.71	- 0.345	0.115
Vitamin A (mcg/ml)	0.42	0.16	- 0.019	0.928
Vitamin E (mcg/ml)	7.60	3.86	- 0.045	0.836
Folate (ng/ml)	10.23	4.46	0.149	0.497
Potassium (mEq/24 hr)	55.16	26.11	0.476	0.025

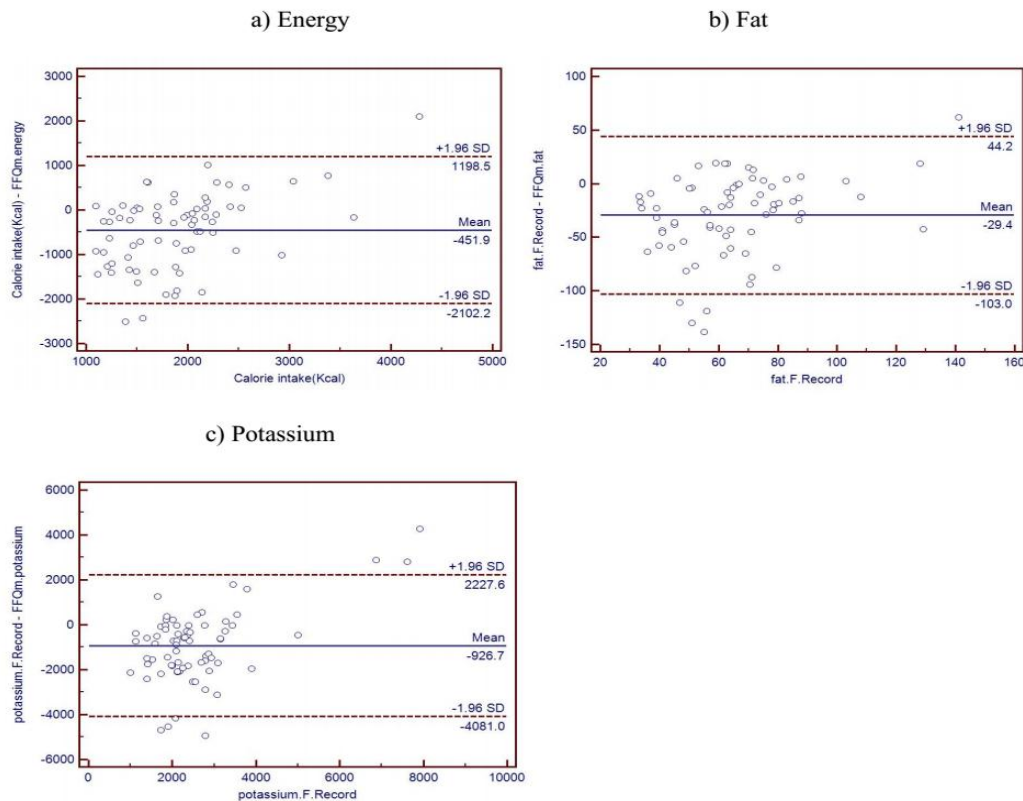


Figure 2. Bland-Altman plots illustrating the relationship between average intake of a) energy and b) fat, and c) potassium estimated using food frequency questionnaire (FFQ) and three-day food record methods (n= 71). The X axis is the amount of intake measured by the reference method (three-day food record).The Y axis is the difference between data (energy, fat, and potassium) intake measured by FFQ (average of FFQ1 and FFQ2) and three-day food record (average of the six three-day food records). The central solid horizontal line indicates the mean difference between the two methods, and the solid lines above and below it indicate ± 1.96 standard deviation (SD).

Discussion

The results of the present study showed a reasonable validity, based on the Bland-Altman plots and correlation coefficients, for energy, fat, and potassium intake evaluation, and good reproducibility of the FFQ over a 6-month period. The observed differences in the dietary intake estimates of some nutrients, such as fat, vitamin A,

and vitamin E, by each of two FFQs and the three-day food records may be due to the use of a non-domestic software (Nutriscan software) for analyzing the three-day food records, whereas the FFQs were analyzed by a domestic software which was designed particularly for this study. Because of the poor cooperation from the participants, only 113 subjects out of 210 completed the project, and this was a limitation of the present

study. Most of the similar studies were completed in a 1-year period, but this study was completed in a 6-month period. Since dietary evaluation during one year results in a complete estimation of the dietary intake and considers the seasonal availability of food items, a 6-month period for this study is another limitation. Since people's dietary intakes, especially the fruit and vegetable groups, varies in different seasons, for reducing the effects of seasonal variations in dietary intakes, we chose autumn and winter seasons since the food consumption in these seasons is relatively similar. Another limitation of the present study was the financial limitation for measuring biomarkers, which obliged us to reduce the sample size. We didn't consider social characteristics, smoking, and BMI in our study, and this accounts for another limitation of the study. Only a few FFQ validity studies (15, 16) have considered subgroups other than gender, like BMI groups. For these studies, a sample size that is large enough to ascertain the differences among subgroup is required.

Previous FFQs that have been designed for the Iranian population consisted of specific kinds of foods and for certain types of diseases, such as diabetes, cardiovascular diseases, and esophageal cancers, but the FFQ in our study included all kind of foods (Traditional, Western, etc.) that had a high frequency of use. So, a complete evaluation of Iranian individuals' food intakes was possible with the present FFQ. As mentioned previously, the amount of energy and macronutrients of traditional foods was determined accurately; hence, the evaluation of food intakes by this FFQ would be precise. Lack of knowledge about the portion sizes and servings of food is one of the common problems of dietary assessment. Because similar dishes have different sizes, for illustrating the determined average uses, an album consisting of 10 photos related to average uses was placed at the beginning of the FFQ. Analyzing the FFQs by multi-function software that was designed particularly for this study decreased the biases compared with that analyzed by non-domestic software, and also, compared to recording data manually.

Using the food record as the reference method is another strength of our study because the food record method provides several unique merits. This method is based on actual intake and may be used to estimate the absolute, rather than relative, intake of energy and other food components such as the macronutrients and some vitamins and minerals that are broadly distributed within the food supply. Also, because the method is open-ended, it can adopt any food or food combination reported by the subject, and they allow an unlimited level of specificity regarding the type of food, food source, food processing method, food preparation, and other details related to describing foods and amounts. Some previous studies used food records as the reference method (17, 18), and other studies used dietary recalls (19-23). Regardless of the kind of reference method, under- and overestimation biases might impact any of the methods used in validation studies (9). The food record method is limited by the fact that a single day of intake is unlikely to be representative of usual individual intake, because day-to-day intake is highly variable for many individuals, so for reducing the adverse impact of this limitation, we used three-day food records which were completed on consecutive days including one weekend day. We also used biomarkers from urine and blood samples as part of our validation study, as errors in dietary methods are not associated with biomarkers.

This study was the fourth FFQ validation study in Iran; the first one was accomplished in Golestan, a province in the north of Iran, as part of the Golestan cohort study of esophageal cancer (24). In the Golestan study, the range of correlation coefficients between the dietary recalls and the FFQ was 0.49–0.82, and the range of intra-class correlation between four FFQs was 0.66–0.89; the correlation coefficients between serum biomarkers and estimated intakes based on four FFQs ranged from 0.06 to 0.37. In the second FFQ validation study, the Tehran Lipid and Glucose Study (TLGS) (9), the correlation coefficients between the dietary recalls and the second FFQ ranged from 0.11 to 0.71, and the intra-class correlation among the two FFQs

ranged from 0.33 to 0.87. The third study was conducted by the National Nutrition and Food Technology Research Institute, Shahid Beheshti University of Medical Sciences, to assess the folate intake status in breast cancer patients (25), and the correlation coefficients between the folate serum levels and the FFQ ranged from 0.17 to 0.44. The correlation coefficients between the means of the two FFQs and three-day food records in our study were lower than the results of Golestan study (24), TLG study (9), McNaughton *et al.* study (26), and Willett *et al.* study (4). The correlation coefficients between the means of the two FFQs and biochemical markers (the average of two measurements) in our study were lower than the results of similar biomarkers in Golestan study (24), Pirouzpanah *et al.* study (25), McNaughton *et al.* study (26), and Willett *et al.* study (4). The correlation coefficient for potassium in our study was higher than that of Day *et al.* study (27). It seems that these differences were observed because of poor cooperation of the participants and few subjects available for final analysis. The Bland-Altman analyses showed fairly good agreement between the average of the FFQs and 3-day food records for energy, fat, and potassium intakes (14) due to the limits of agreement being approximately equal to two SD of the food records data. The results of another FFQ validation study (28), in which the Bland-Altman method was used, showed fairly good agreement between the average of the FFQs and the average of the 24-hour dietary recall for energy, fat, protein, fiber, and carbohydrate intakes, and this is in accordance with our study. A similar finding for energy intake was reported by Macedo-Ojeda *et al.* (29). Compared with the dietary record, the FFQ2 overestimated intakes for energy, and the Bland-Altman analyses showed fairly good agreement between the FFQ2 and the average of the dietary records for energy. In the present study, the differences between the dietary intake from FFQ1 and FFQ2 were not significant, except for vitamin A. The higher correlation among men in our study was similar to the findings in TLGS (9) and Ocke *et al.* study (30). In future studies, for better evaluation of the validity and reproducibility of the FFQ, it would be better to accomplish the project in a 1-year period; thereby the dietary intake of the

population in all seasons will be assessed. For transferring blood and urine samples from several cities to the destination city more safely and cost-effectively, it is better to use special shipment services instead of using domestic flights.

Conclusion

According to the results of this study which used an integration of two FFQs, three-day food record, and biomarkers in serum and urine samples, the FFQ has reasonable validity for energy, fat, and potassium intake evaluation and good reproducibility over a 6-month period. Since the samples of this study were selected from different provinces of Iran, we can generalize the results of the study to the entire country. So, the validated FFQ seems to be a reasonable tool for evaluating nutrient intakes in Iranian urban population..

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