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Lora, Karina R.; Lewis, Nancy M.; Eskridge, Kent M.; Stanek-Krogstrand, Kaye; and Ritter-Gooder, Paula K., "Validity and Reliability of an Omega-3 Fatty Acid Food Frequency Questionnaire for First-Generation Midwestern Latinas" (2010). *Nutrition and Health Sciences -- Faculty Publications*. 9. <https://digitalcommons.unl.edu/nutritionfacpub/9>

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# Validity and Reliability of an Omega-3 Fatty Acid Food Frequency Questionnaire for First-Generation Midwestern Latinas

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K. R. Lora was a graduate student at the University of Nebraska–Lincoln at the time this project was conducted.

## Abstract

This study tested the hypothesis that a culturally developed omega-3 (n-3) fatty acid food frequency questionnaire (FFQ) could be an accurate instrument to capture n-3 fatty acid food intakes of first-generation Midwestern Latinas. The goal of the study was to assess validity and test-retest reliability of an FFQ to estimate total n-3 fatty acid (total n-3), *n*-linolenic acid (ALA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA) intakes. An n-3 FFQ was developed and pilot tested. Two FFQs and 3 nonconsecutive 24-hour recalls were collected from 162 participants. Pearson correlation and paired *t* test were used to test the hypothesis. Correlation of the 2 FFQs was 0.71 for total n-3, 0.65 for ALA, 0.74 for EPA, and 0.54 for DHA ( $P < .01$ ). The means of the 2 FFQs and of the 24-hour recalls were not significantly different for total n-3 and ALA ( $P > .05$ ), but were significantly different for EPA and DHA. The n-3 FFQ had acceptable reliability, validated only total n-3 and ALA, and provided relevant findings about the n-3 eating habits of Midwestern Latinas.

**Keywords:** omega-3 fatty acids, food frequency questionnaire, Hispanic Americans, reproducibility of results, validation study, *n*-linolenic acid, eicosapentaenoic acid, docosahexaenoic acid

**Abbreviations:** ALA, *n*-linolenic acid; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; FFQ, food frequency questionnaire; LC PUFA, long-chain polyunsaturated fatty acid; n-3, omega-3; NCC, Nutrition Coordinating Center; NDSR, Nutrition Data System for Research; USDA, United States Department of Agriculture

## 1. Introduction

The omega-3 (n-3) fatty acids (*n*-linolenic acid [ALA], eicosapentaenoic acid [EPA], and docosahexaenoic acid [DHA]) are essential for different physiologic processes in humans [1–3]. Eicosapentaenoic acid and DHA are part of membrane phospholipids and can influence the fluidity, thickness, and deformability properties of the membranes. Eicosapentaenoic acid and DHA are particularly abundant in the myocardium, retina, and brain and are important for adequate functioning of these tis-

ues through the life cycle, modulating many physiologic processes [1, 3]. Epidemiologic and interventional studies have demonstrated the beneficial effects of n-3 fatty acids on cardiovascular disease [4–6]. Flaxseeds, walnuts, and canola oil are food sources to achieve recommended ALA intakes, whereas consumption of 2 fatty-fish meals per week is recommended to meet EPA and DHA recommendations [7]. Greater benefit on cardiovascular health has been attributed to food sources of EPA and DHA compared with ALA, although controversy still exists [1, 7, 8].

Heart disease accounts for 24% of the deaths in Latinos living in the United States [9]. It has been reported that, in the United States, Latinas have greater risk for heart disease and stroke risk factors [10] compared with white and Asian women. Reported intakes (in grams per day) of ALA, EPA, and DHA of Mexican American women 20 to 49 years old are in the range of 1.1 to 1.2, 0.02 to 0.03, and 0.05 to 0.06, respectively [11]. Higher intakes (in grams per day) of 1.3 for ALA and in the range of 0.03 to 0.04 and 0.06 to 0.07 for EPA and DHA, respectively, have been estimated for the US women population 20 to 59 years old [12]. Low intakes of EPA and DHA found mainly in marine sources may explain low intakes in Latinos as observed in the frequency of fish consumption of  $0.3 \pm 0.2$  times per day of Latinas in Connecticut [13].

Food frequency questionnaires (FFQs) are often used to measure the association of diet and disease [14]. Food frequency questionnaires have become widely used because of their feasibility to assess intake over a long period, reduce error introduced from day-to-day variability, and decrease respondent burden as compared with other dietary assessment methods [15]. Two FFQs to estimate n-3 fatty acid intakes have been validated previously. An Australian FFQ that estimates long-chain n-3 polyunsaturated fatty acids (LC n-3 PUFAs) reported Spearman correlations coefficients of 0.75, 0.64, 0.62, and 0.72 for total LC n-3 fatty acids (EPA, docosapentaenoic acid, and DHA fatty acids, respectively) when tested against 3 food records [16]. An FFQ validated against three 24-hour recalls as reference method to estimate n-3 fatty acid intakes in cardiovascular patients in the US Midwest [17] reported Pearson correlation coefficient of 0.42.

The lower n-3 fatty acid intake of Latinos living in the United States could be in part explained by the use of dietary instruments that do not adequately capture the n-3 fatty acid content of the diets of Latinos. Thus, dietary instruments developed to estimate n-3 fatty acid intake in the foods normally consumed by Latinos are important because of their higher sensitivity to changes in diet. There is good evidence that an FFQ can measure dietary change [18].

Dietary intakes of n-3 fatty acids and marine food consumption, sources of EPA and DHA of white persons living in the US Midwest, are reported to be low [17, 19–21]. Cardiac patients, physically active adults, and low-income US Midwestern pregnant women reported intakes of 0.18 to 10.15,  $0.887 \pm 0.121$ , and  $1.060 \pm 0.030$  g/d, respectively [17, 21, 22]. Adequate intakes for ALA for women at least 19 years old have been set at 1.1 g/d [2], and a combined minimum intake of 0.5 g/d EPA and DHA for cardiovascular health has been recommended [23]. Little is known of the n-3 fatty acid intakes of Latinos living in the United States as measured by FFQs that include cultural foods that are part of the Latino diet.

Latinos will comprise 15.8% of the population in the US by 2015 [24]. Minority women, such as Latinas, have

a greater chance of experiencing heart disease [10] and lower awareness of heart disease as their leading cause of death compared with white women [25]. There is a need for culturally appropriate validated dietary instruments to measure dietary intake and use in nutrition interventions in ethnic/racial groups in a nation that is becoming more racially and ethnically diverse. The present study aimed to test the hypothesis that a culturally developed n-3 FFQ could be an accurate instrument to capture n-3 fatty acid food intakes of first-generation Midwestern Latinas. The objectives were (1) to develop an FFQ that could be used for evaluating n-3 fatty acid food intakes of first-generation Midwestern Latinas and (2) to assess the concurrent validity and test-retest reliability of the FFQ against 24-hour recalls to estimate total n-3, ALA, EPA, and DHA intakes in the same population.

## 2. Methods and materials

Several stages were used to develop and test the n-3 FFQ for validity and reliability.

### 2.1. FFQ development

Several steps were conducted in this stage to develop a culturally appropriate dietary instrument using a pilot FFQ validated for estimating total n-3 fatty acid intakes in cardiac patients [17] in our laboratory. The purpose of the developmental work was to conduct sequential steps in an attempt to obtain an accurate dietary instrument intended to capture n-3 fatty acid food intakes of first-generation Midwestern Latinas. The stages of development were as follows.

#### 2.1.1. Generation of a list of major n-3 fatty acid food contributors

A convenience sample of 5 first-generation Latinas was interviewed to obtain preliminary dietary information to describe the major food contributors of n-3 fatty acids in their diet and to obtain recipes for culturally specific prepared dishes and other prepared foods and the respective portion sizes that were part of their daily diet. In this stage, participants each completed a single 24-hour recall. In the 24-hour recall method, subjects are asked to recall the subject's exact food intake during the previous 24-hour period. This method assesses the actual intake of individuals and provides a valid measure of the intake of a group or population [14]. Additional information obtained were foods and meals that were part of their daily diet, foods that they prepared for themselves, groceries purchased and the markets used, food ordered when dining out, and foods used in their diet from any supplemental food programs. In addition, when culturally specific prepared dishes (ie, enchiladas, tacos dorados) were reported, recipe information was obtained. When preparation methods of cultural dishes varied among participants, all recipes were examined to select a standard recipe for nutrient analysis and assignment of values for the FFQ. A final step was to conduct a literature review [26–

30] to include reported foods eaten by Latinos and that could be sources of n-3 fatty acids. This listing of n-3 food contributors to the diets was used to modify the n-3 FFQ for use in the population.

### 2.1.2. Definition of portion sizes and database development

A standard or medium portion size for single food items reported on the preliminary interviews was defined using the US Department of Agriculture (USDA) MyPyramid daily recommendation guidelines [31]. Portion sizes for the culturally specific dishes were determined using the recipe yield and number of individuals served. Foods in the FFQ were grouped as meats, fish, seafood, eggs, dairy products, vegetables, fruits, breads and cereals, snacks, oils and fats, nuts and seeds, beans, and prepared dishes. Each food item line referenced a single food and was not a composite of several like items. For example, all fish species were individually listed, including salmon and tuna.

Food composition data were obtained from the Nutrition Data System for Research (NDSR) 2007 [32], a dietary analysis program that uses the Nutrition Coordinating Center (NCC) Food and Nutrient Database. The primary source of nutrient values and nutrient composition of the NCC is the USDA Nutrient Data Laboratory, which lists more than 18 000 foods and 7 000 brand products. Composition data are derived from independent analysis, food manufacturers, scientific literature, published food tables, and unpublished analytical data [33, 34]. Imputed values in the NCC are obtained from using values of a different but similar food, different forms of the same food, and household recipes or commercial product formulation for multi-component foods, converting values from nutrient label information of commercial food products or assuming a zero value. It was possible to analyze recipes using ingredients and preparation methods for dishes frequently consumed by the population studied but not yet in the database. The NDSR 2007 was chosen because of its extensive list of analytical and imputed values, including those for ethnic Latino dishes, providing a comprehensive nutrient analysis program for this FFQ study.

Each reported food and culturally specific dish was analyzed for total n-3 fatty acid, ALA, EPA, and DHA content in grams per medium serving. In addition, foods from the original FFQ [17] were analyzed for ALA, EPA, and DHA content, as this FFQ was only designed to assess total n-3 intake. Food composition analyses were conducted using NDSR 2007 [32]. The resulting nutrient analysis was used to rank single foods (ie, canola oil, walnuts, pinto beans) that contributed at least 0.010 g total n-3 fatty acid per medium serving. Because the food composition of prepared dishes (ethnic dishes included) was made of several ingredients, all prepared dishes contained at least 0.010 g total n-3 fatty acid per medium serving. Several foods consumed by the Latino cultural group were included in the list (Mexican queso fresco, corn tortilla, bolillo).

### 2.1.3. Pilot testing of the n-3 FFQ

The resulting preliminary FFQ was analyzed first for content validity with 3 nutrition experts. Methodology used in content validity involves a panel of experts who evaluate the instrument by examining the plan and procedures used in constructing the instrument, as well as objectives of the instruments, content areas, and the level of difficulty of the questions [35]. Next, the FFQ was pilot tested in a convenience sample of 10 first-generation Latina women to evaluate its effectiveness and to obtain participants' usual frequency of consumption of each food from the preliminary list of foods [36]. Food models were used to facilitate recall and to help estimate portion size. [14]. Participants provided feedback on the instructions, understanding of portion size and frequency of food consumption, and readability of the FFQ. An open-ended question – "Are there any other foods that you eat at least once a month?" – was asked to include missing foods. After changes were made, the n-3 FFQ contained 183 food items and 26 prepared dishes (15 culturally specific dishes). The n-3 FFQ was translated and back-translated to and from Spanish (KL).

## 2.2. Sociodemographic questionnaire

A sociodemographic questionnaire adapted from the Behavioral Risk Factor Surveillance System 2006 Questionnaire, Spanish version [37], was used to obtain information regarding age, country of birth, ethnicity, years of schooling, annual household income, language spoken at home, employment status, age when first came to the United States, and years of permanency in the United States for each participant.

## 2.3. Sample and study design

One-hundred sixty-two first-generation Latinas from the cities of Lincoln and Omaha in Nebraska served as participants in the present study. Because nutrient intake varies by sex, contributing to between-subject variation, as well as the fact that minority women have an increased risk for heart disease, we decided to sample only women. Women were recruited from Latino community centers, churches, and physical activity community service project sites. Convenience and snowball sampling was used for recruitment [38]. Participants were between the ages of 20 to 50 years, were not pregnant or lactating at the time of the study, and reported no current illness or smoking. Only one participant was recruited from each household or family. One-on-one interviews were scheduled in advance to accommodate participant schedules and were conducted between October 2007 and August 2008 by a trained bicultural interviewer at the participant's home, church, community center, or community project site. One-on-one interviewing allows data collection for all questions, leading to a high response rate and low measurement error [35]. The same interviewer conducted all interviews to minimize variations in protocol and to provide quality con-



trol across all subjects. Community health workers and social workers served as liaisons. Sociodemographic information was obtained during the first interview. Interviews took approximately 1 hour to complete and were conducted in Spanish. Participants who completed the study received a \$30 gift card as compensation for their participation. The Institutional Review Board for the Protection of Human Subjects at the University of Nebraska-Lincoln approved the study protocol, and all participants provided written informed consent.

#### 2.4. Data collection methodology and assessment

Two FFQs, 4 weeks apart, and 3 nonconsecutive 24-hour recalls (including a weekend day) within the 4-week period were collected. The 24-hour recall was used as the reference method [14, 36]. Participants met with the interviewer to complete one FFQ and one 24-hour recall in each meeting at the beginning and the end of the 4 weeks and provided one announced 24-hour recall by phone in the interim. To complete the FFQ, the following protocol was used: (1) The interviewer read each food item and mixed dishes from the list of foods. (2) If the participant ate the food, then she was asked to indicate whether her usual serving was small, medium, or large. Visual aids (food models, pictures, kitchen utensils) were used to estimate serving sizes. (3) Participant was asked to recall how often she ate the food over the past month: none, once a month, 2 to 3 times a month, 1 to 2 times a week, 3 to 4 times a week, 5 to 6 times a week, and daily (once or twice a day) [39]. The food recall was obtained for each participant using the USDA 5-Step Multiple Pass Method [40]. Participants were queried in 5 steps. (1) Quick list. In this step, a quick list of foods and beverages consumed was collected. (2) The forgotten foods list. An inquiry was made into categories of foods that have been documented as frequently forgotten, such as crackers or nuts. (3) The time and occasion at which foods were consumed. (4) The detail cycle, which elicits descriptions of foods and amounts eaten aided by the use of the food models and measuring guides. (5) The final probe. This step provides the final check for completeness of the food recall [40]. During the first meeting interview, visual aids (food models, pictures, kitchen utensils) were used to instruct participants on estimating serving sizes when reporting their intake by phone [14].

A template developed in Microsoft Excel 2003 (Microsoft Corp, Seattle, WA) containing nutrient database for total n-3, ALA, EPA, and DHA per medium serving for each single food and prepared dish was used to compute nutrients intake in the FFQ. N-3 fatty acids food composition of foods in the n-3 FFQ was obtained from the NDSR 2007 program and imputed in the Microsoft Excel template to calculate n-3 fatty acids for each participant. In the template, the selected list of foods was incorporated in an FFQ format; and serving sizes (small, 0.5; medium, 1; and large, 1.5) were defined for each food item [31]. Frequency of consumption was

classified as none, once a month, 2 to 3 times a month, 1 to 2 times a week, 3 to 4 times a week, 5 to 6 times a week, and daily (once or twice a day). In this template, estimates of intakes of total n-3 and each n-3 fatty acid (ALA, EPA, and DHA) in the FFQ were calculated for each single food item and prepared dish by multiplying the nutrient value for each food by the frequency of consumption times by the amount of that food eaten for the respective n-3 fatty acid (ALA, EPA, and DHA) content of the food. When the participant ate a smaller or larger portion than the defined medium portion size, the amount entered was calculated based on the portion size for that food in the n-3 FFQ. Twenty-four hour recalls were analyzed using the NDSR 2007. Key verification method was used as quality control for data entry.

#### 2.5. Statistical analyses

Descriptive statistics for sociodemographic data and n-3 fatty acid dietary intake were estimated for the FFQs and the 24-hour recalls. Dietary data were log-transformed (corrected) to improve the normality of the distribution with the formula  $\log(x + 1)$  because intake values could be zero [41] before computing calculations. For reliability, Pearson correlation coefficients were calculated to compare the association between the first and second administration of the FFQ by total n-3, ALA, EPA, and DHA in a test-retest approach. To assess concurrent validity, corrected and adjusted Pearson correlation coefficients and paired *t* tests were computed between the mean of the 2 FFQs and the mean of the three 24-hour recalls. To account for random errors and within-person variability, the variance ratio (ratio of within-subject [ $s_w^2$ ] to the between-subject [ $s_b^2$ ] variation) was calculated to deattenuate (adjust) the correlations of the 24-hour recalls [14, 36, 42, 43]. Because large within-subject variation in intake lowers and makes correlations between the reference and test method less significant, this effect can be amended by deattenuation (adjusting) the correlation coefficient [14] with a resultant increase in the correlation coefficients. The *P* value for the deattenuated correlation coefficients was calculated according to Rosner and Willett [44]. With 162 respondents, power was approximately 90% for (1) detecting a correlation coefficient of 0.2 or larger and (2) detecting a difference of at least 25% of the size of the standard deviation of a difference for the paired *t* test [45]. Frequencies, means, standard deviations, correlations, and *P* values are presented in the text and in tabular form. The level of significance was set at  $\alpha = .05$ . Statistical analysis was conducted using SAS (version 9.1, 2006; SAS Institute, Inc, Cary, NC).

### 3. Results

All 162 participants completed the study. Mean  $\pm$  SD age of participants was  $34.3 \pm 8.2$  years, with nearly 40% of participants in the 20- to 30-year range. More than two thirds (68%) of the women were from Mexico, 61% had

**Table 1.** Total n-3 PUFA, ALA, EPA, and DHA intakes (in grams per day)<sup>a</sup> of Latinas (n = 162) living in the US Midwest as estimated by n-3 FFQ<sup>b</sup> and 24-hour recalls<sup>c</sup>

	FFQ1	FFQ2	Mean FFQ	24-h recall 1	24-h recall 2	24-h recall 3	Mean 24-h recalls
Total n-3 PUFA	1.42 ± 0.91	1.06 ± 0.65	1.22 ± 0.70	1.20 ± 1.10	1.28 ± 1.17	1.25 ± 0.97	1.23 ± 0.78
ALA	1.27 ± 0.87	0.94 ± 0.60	1.10 ± 0.65	1.13 ± 1.06	1.13 ± 1.05	1.19 ± 0.97	1.14 ± 0.74
EPA	0.04 ± 0.04	0.03 ± 0.04	0.04 ± 0.04	0.02 ± 0.06	0.05 ± 0.14	0.01 ± 0.03	0.03 ± 0.05
DHA	0.09 ± 0.08	0.07 ± 0.07	0.08 ± 0.06	0.04 ± 0.10	0.07 ± 0.25	0.03 ± 0.05	0.05 ± 0.09

<sup>a</sup> Values are means ± SD.

<sup>b</sup> Administered 4 weeks apart.

<sup>c</sup> Collected on nonconsecutive days, including 1 weekend day and in between administration of the FFQs.

completed high school, and 43% had annual incomes in the range of \$10 000 to \$20 000. Most women spoke Spanish at home (82%), had a mean age of arrival to the United States of 25.1 ± 9.0 years, and had lived in the United States at the time of the study for 9.3 ± 6.4 years.

Means ± SDs for each replicate of the FFQ and 24-hour recall and for the mean of both dietary methods are shown in Table 1. The n-3 FFQ estimated 99% of the intake of total n-3 from the recalls and 96% from ALA,

and gave a higher estimate of EPA and DHA intakes by 133% and 160%, respectively. Mean ALA intake assessed by the n-3 FFQ accounted for 90% of total n-3 intake. Table 2 shows the rank listing of foods that provided 99% and 43%, respectively, of daily total n-3 from the FFQ and the 24-hour recalls.

The n-3 FFQ had adequate reliability, but low validity. Reliability correlation coefficients in the n-3 FFQ ranged from 0.54 for DHA to 0.71 for total n-3. Validity correlation coefficients were only corrected (log-transformed to improve normality) when performing deattenuation calculations presented difficulties. First, the variance ratios were 2.13 and 2.01 for total n-3 and ALA, respectively, whereas the variance ratios were 203 for EPA and zero for DHA. The within-subject variation was high, whereas the between-subject variation was low, in the nutrients studied. Eicosapentaenoic acid and DHA showed highest within-subject variation and near-zero between-subject variation, which made the deattenuation calculations unrealistic. Second, although the deattenuated correlation coefficients were calculated for these 2 nutrients (0.42 for total n-3 and 0.44 for ALA), the *P* value of the deattenuated correlation coefficients for total n-3 and ALA was not significant (*P* = .08). This finding could be explained by the extra variability added when the corrected correlation coefficients were adjusted. Thus, Pearson correlation coefficients were 0.32, 0.34, 0.28, and 0.24 for total, n-3, ALA, EPA, and DHA, respectively. The paired *t* test showed no significant differences (*P* > .05) between the n-3 FFQ and the 24-hour recalls for total n-3 and ALA, implying that there was no detectable bias. However, EPA and DHA showed significant differences (Table 3).

**Table 2.** Rank order listing of foods that contributed to daily total n-3 PUFA intake estimated by the n-3 FFQ<sup>a</sup> and the 24-hour recalls<sup>b</sup> in Latinas living in the US Midwest (n = 162)

Rank order	Food		% Each food provided to total n-3 PUFA intake	
	FFQ	24-h recall	FFQ	24-h recall
1	Vegetable oil <sup>c</sup>	Vegetable oil	16	11
2	Canola oil	Canola oil	13	7
3	Mayonnaise, regular	Mayonnaise, regular	8	4
4	English walnuts	Flax seeds	7	3
5	Beef steak	Salad dressing "Ranch type"	7	3
6	Pinto beans	Whole milk	6	3
7	Salad dressing "Ranch type"	Pinto beans	6	2
8	Fresh or frozen salmon	Beef cuts	5	2
9	Whole milk	Cookies and bars	5	2
10	Chicken burger, breaded	Soy milk	5	1
11	Beef cuts	Corn oil	5	1
12	Corn tortilla	Tilapia	4	1
13	Chicken leg, no skin	Fresh or frozen salmon	4	1
14	Tilapia	Tres leches pastel <sup>d</sup>	4	1
15	Shrimp	Corn tortilla	4	1

<sup>a</sup> Intake assessed by the mean of the 2 FFQs contributing to 99% of daily total n-3 PUFA intake.

<sup>b</sup> Intake assessed by the mean of the three 24-hour recalls contributing to 43% of daily total n-3 PUFA intake.

<sup>c</sup> Commercial mixture, other than canola.

<sup>d</sup> Dessert made of milk, pound cake, and sugar.

**Table 3.** Reliability and validity estimations of an n-3 FFQ measuring total n-3 PUFA, ALA, EPA, and DHA of Latino women living in the Midwest

	Reliability		Validity	
	Pearson correlation FFQ1 vs FFQ2	Pearson correlation FFQ vs 24-h recalls	Paired <i>t</i> test	
Total n-3 PUFA	0.709†	0.324*	0.7369 <sup>a</sup>	
ALA	0.654†	0.337*	0.6856 <sup>a</sup>	
EPA	0.740†	0.275*	0.0127*	
DHA	0.537†	0.236*	<0.0001*	

<sup>a</sup> Not significant ; \* *P* < .05 ; † *P* < .01.

#### 4. Discussion

In the present study, we developed and assessed the test-retest reliability and concurrent validity of a culturally appropriate FFQ that estimates total n-3, ALA, EPA, and DHA intakes in first-generation US Midwestern Latinas.

The FFQ demonstrated acceptable reliability for all LC PUFAs measured. Correlations of 0.54 to 0.71 were similar to values (0.5-0.8) found in FFQs measuring other nutrients and in other populations [14, 36, 46]. However, higher n-3 FFQ reliability correlations (0.83-0.90) have been reported in other studies [16, 17]. The greater variability in food intakes reported in women compared with men [42] and the limited availability of marine dietary sources of n-3 fatty acids in this geographical area, affecting habitual intake, may explain the lower values found in this study.

The n-3 FFQ was validated for estimating total n-3 and ALA dietary intakes. The n-3 FFQ appears to accurately capture total n-3 and ALA intake reported by 24-hour recalls. Total n-3 and ALA estimates showed non-significant mean differences between n-3 FFQ and 24-hour recalls and had similar low variance ratios. Validity of the n-3 FFQ for estimating total n-3 in the present study was lower than that obtained in a pilot study of a 152-item n-3 FFQ (0.32 vs 0.42) [17].

The n-3 FFQ was not validated for estimating EPA and DHA intakes when compared with 24-hour recalls. Both fatty acids showed statistically significant differences ( $P < .05$ ) between the 2 measurement methods and had higher variance ratios. The attempt to deattenuate their correlation coefficients showed a nonsignificant  $P$  value, implying that extra variability was added. Within- and between-subject variance is a function of the nutrient of interest [47]. Infrequent consumption of foods with high concentrations of nutrients, such as n-3 fatty acids, often has high within-subject variation, complicating accurate estimates of usual intake. Women, compared with men, have higher within-subject and lower between-subject variability for several nutrients, especially for fat, because of inconsistent use of low-fat products or decreased intake of fat-containing products [46]. Sex is a major contributor to the sources of variance in 24-hour recalls for different nutrients. Women reported higher mean intakes of PUFA on weekends compared with weekdays and had higher variance ratios than men for PUFA when compared with other nutrients [42]. A possible explanation is that EPA and DHA intakes had a high within-subject and low subject variability that affected their correlation coefficients, lowering them and making the deattenuation calculations unrealistic.

Other FFQs have been validated for measuring EPA and DHA. Sullivan et al [16] obtained Spearman correlation coefficients of 0.64 and 0.72 for EPA and DHA, respectively, when they compared a 28-item FFQ that estimated LC n-3 PUFAs with 3 weighed food records

in Australians. Pearson correlation coefficients of 0.59 and 0.55 for EPA and DHA have been reported when comparing a 129-food item FFQ with 7-day weighed records designed to measure dietary fatty acid in the United Kingdom [48]. The higher correlation coefficients obtained for EPA and DHA in these studies compared with the present study could be attributed to the population studied and the use of food records instead of 24-hour recalls. The subjects of the Australian and UK studies were highly educated university staff and students; and a considerably shorter FFQ was tested, possibly influencing respondent burden [14].

Food records are considered a more appropriate reference method to validate an FFQ [14]. The present study used 24-hour recalls because of ease of collection and usefulness in estimating intakes in culturally specific populations among individuals of low literacy [14, 49, 50]. Several FFQ validation studies involving Latino populations have used the 24-hour recall as the reference method [26, 43, 51, 52]. Although we collected 3 nonconsecutive 24-hour recalls in 1 month, we found that the within-subjects variance was high for the nutrients studied, especially for EPA and DHA, indicating that more measurement days were needed to control for variation.

Food frequency questionnaires are retrospective methods that assess usual and long-term dietary intakes of individuals. Because of the high variability in intake of nutrients that are found in high concentration in few foods such as the n-3 fatty acids, it was not unexpected that the food recalls showed lower mean EPA and DHA intakes compared with the n-3 FFQ, implying that the recalls were not likely to account for overall n-3 fatty acids foods. In that regard, FFQs provide a better understanding of long-term consumption.

The top 5 foods sources of total n-3 intakes as estimated from the FFQ were vegetable (other than canola) and canola oil, regular mayonnaise, walnuts, and beef. Similarly, the top 5 n-3 fatty acid food sources for the 24-hour recalls were vegetable (other than canola) and canola oil, regular mayonnaise, flax seeds, and salad dressing "Ranch type."  $\alpha$ -Linolenic acid is the most abundant n-3 fatty acid in foods of vegetable origin and thus the largest contributor to total n-3 estimates. In the present study, mean ALA intake as assessed by the n-3 FFQ accounted for 90% of total n-3 intake, illustrating that the principal food sources of total n-3 were likely to be the sources of ALA. Although the FFQ contained many cultural foods, pinto beans, corn tortilla, and shrimp, foods frequently eaten by Latino subgroups, contributed the most to total n-3 intakes in the participants. Food sources of EPA and DHA such as beef, fish, chicken, and shrimp contributed up to 7% of total n-3 and are in agreement with data from the Continuing Survey of Food Intakes by Individuals II in which meat, poultry, and fish account for most EPA and DHA in women older than 20 years [53].



The present study had limitations. The number of replicates of the reference method collected may have not been enough for the type of nutrient under study. Our population was hard-to-reach, highly mobile immigrants; and collection of more than three 24-hour recalls would have been difficult to obtain. It would be of interest to assess validity of the n-3 FFQ without the addition of mixed dishes to estimate if changes in validity would result. Biomarkers measuring n-3 dietary intakes were not collected. In addition, the low overall intake of participants that affected the within- and between-subject variation could have affected intake estimates. In further studies, a comparison between interviewer vs self-administration of the n-3 FFQ to examine the effect of data collection on reliability and validity of the questionnaire would be of interest. Further work with the n-3 FFQ needs to take into account longer periods between administration of the FFQ and reference method to elucidate if timing is a factor that may affect validity.

In conclusion, we accepted the hypothesis that the n-3 FFQ could be an accurate instrument to capture n-3 fatty acid food intakes of first-generation Midwestern Latinas. The n-3 FFQ had adequate reliability and validated total n-3 and ALA, but not EPA or DHA. Future studies on the development of culturally appropriate n-3 fatty acids dietary assessment methods would benefit from larger validation studies that could include several replicate measurements due to the high variability of the nutrients studied.

**Acknowledgments** — Funding for this study was provided by the Vice-Chancellor for Research Minority Grants at the University of Nebraska. Special thanks to Daryl A. Travnicek for his statistical assistance and to Paula Ritter-Gooder for her editorial assistance. We are grateful to Diana Garcia, Blanca Follwer, Gabriela Ortiz, and July Braunsroth for their assistance during recruitment.

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