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Original Research Article

Assessing vitamin A intake among pregnant women: The creation and validation of a semi-quantitative FFQ specific for vitamin A in a tertiary health care center

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A B S T R A C T

Background: This study aims to develop a semi-quantitative Food Frequency Questionnaire (FFQ) to assess vitamin A levels using 24-hour food recall in pregnant women and further validate the formulated FFQ. Our study found a significant correlation between the 24-hour recall and FFQ results, indicating that the developed FFQ is a valid and useful tool for assessing vitamin A intake in pregnant women.
Materials and Methods: This was a two-phase study conducted at a Tertiary health care centre in Chennai, Tamil Nadu, consisting of two groups: Group A (55 participants) and Group B (68 participants). Phase I consisted of administering a 24-hour food recall to group A participants to collect their vitamin A intake and develop a FFQ. During Phase II of the study, FFQ was administered to Group B participants, which estimated vitamin A intake over the past 30 days. These patients were then contacted within a week of the administration of FFQ, and a 24-hour recall was administered. The results collected from the 24-hour recall were used to validate FFQ.
Results: Results showed r value under Pearson coefficient was 0.743 (95% CI = 0.613 - 0.834, p <0.001), indicating a strong and significant relationship, and rho under Spearman coefficient was 0.686 (95% CI = 0.535 - 0.794, p <0.001) indicated the presence of a significant correlation suggesting the FFQ is valid.
Conclusions: The results of this study found that the developed FFQ is a valid and useful tool for assessing vitamin A intake in pregnant women and can be utilised to assess vitamin A intake.

1. Introduction

Pregnancy is a critical period during which optimal nutrition is essential for both the mother and the developing foetus. Adequate nutrition during pregnancy profoundly affects the health of the mother and the development of the fetus.[1] Inadequate maternal diet has been implicated in not only increased risk of complications during pregnancybut also linked to chronic conditions in childhood.[2] Around 20% to 30% of pregnant women worldwide suffer from some vitamin deficiency.[3]

Micronutrients, which include vitamins and minerals, have a significant impact on the health of both pregnant women and their developing fetus.[4] Folic acid supplementation during preconception and early pregnancy has been shown to prevent 40–80% of neural tube defects.[5] Vitamin B deficiencies, including riboflavin and niacin, have been associated with preeclampsia, heart defects, and low birth weight infants.[6] Maternal vitamin D deficiency has been associated with neonatal rickets[7] and various pregnancy complications, including preeclampsia and gestational diabetes mellitus.[8,9] Similarly, vitamin A is an essential micronutrient during pregnancy, which has various important downstream physiological effects

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for both the mother and the foetus. Vitamin A enhances immune function, which is crucial for both the foetus and the mother. A deficiency can leave them more susceptible to infections.[10] Retinoic acid, an active metabolite of vitamin A, is essential for organogenesis, affecting the development of the heart, eyes, ears, and limbs.[11] Vitamin A is vital for cell division and differentiation, and it helps maintain epithelial cellular integrity, which is crucial for foetal growth.[12] Vitamin A has also been found to be linked to various aspects of iron metabolism. Vitamin A deficiency has been shown to impair iron mobilisation with subsequent accumulation in tissues.[13]

Accurate assessment of nutrient intake, including critical micronutrients such as vitamin A, is crucial during pregnancy, as deficiencies in these nutrients can have adverse pregnancy outcomes.[14] However, the tools used to estimate the vitamin A status are very limited and expensive as it is done via measuring the serum and breast milk retinol concentration.[15] Moreover, there is a significant lag time where the liver's vitamin A reserves need to be dangerously low before the serum retinol concentrations are affected.[15] There is a need for an inexpensive validated tool that can give an estimate of the vitamin A status in pregnant women. Food Frequency Questionnaires (FFQs) have emerged as valuable tools for capturing the habitual dietary patterns of individuals over an extended period[16] and can provide an alternative tool for assessing the vitamin A intake in pregnant women. These can be qualitative or semi-quantitative in nature.[17] Food frequency questionnaires have historically been used to estimate the nutritional status of people.[18]

This study aims to evaluate the daily vitamin A intake of pregnant women using a 24-hour food intake assessment method. Additionally, based on the results obtained from the 24-hour dietary intake assessment, this study aims to develop a semi-quantitative Food Frequency Questionnaire (FFQ) specifically designed to assess vitamin A levels. Subsequently, the FFQ will be validated for its effectiveness in summarising vitamin A intake for a period of 30 days preceding administration of the FFQ. Through this research, we hope to enhance our understanding of pregnant women's vitamin A intake and provide a reliable tool for future assessments in large-scale studies.

2. Materials and Methods

This study was conducted in two phases at a tertiary health care centre in Chennai, Tamil Nadu. The study consisted of two groups, group A and group B. Group A was assessed as a part of an ICMR-approved study with 55 participants, which was conducted in 2018 as Phase 1, and Group B was analysed from July to October 2022 in Phase 2. Group B consists of 68 patients [Figure 2]. The study aimed to examine the dietary habits of healthy pregnant women who came for regular check-ups at the hospital.

A total of 150 women were selected to participate in this study. All these women were in their second trimester and had no known underlying health conditions. They had come for a routine pregnancy visit. Before administering the Semi-quantitative Food Frequency Questionnaire (FFQ) and conducting 24-hour recalls, basic information, including the participants' names, ages, weights, number of previous births (parity), and haemoglobin concentration levels, were collected. Out of the 150 selected, 27 were not included in the final analysis due to not being able to complete the FFQ and the 24-hour recall procedure [Figure 3].

It is important to note that the participants who were included in the Semi-quantitative FFQ and 24-hour recall assessments were those who provided written consent. The consent form was available in both English and Tamil languages to ensure the understanding of all participants.

2.1. *Process of development of FFQ: Phase 1*

We gathered data from 55 pregnant women who had come in for their regular check-ups. We used a 24-hour recall method to collect information about their food intake. From this recall data, a detailed list of food items was created. After consulting the IFCT 2017 guidelines for nutrition released by the NIN and referring to the food atlas developed by the Madras Diabetes Research Foundation, a list of food items with a relevant amount of vitamin A of more than 10ug/100gm RAE was created. To ensure accuracy in food quantity recall, the food atlas provided pictorial representations of each food item along with its corresponding quantity. This was done as a part of an ICMR-STS grant research study.

2.2. *Process of validation of FFQ: Phase 2*

The semi-quantitative FFQ that was formulated in the First part was taken and then administered to 68 women who had come to the obstetrics outpatient department. Along with the semi-quantitative FFQ, contact information was also taken. With the semi-quantitative FFQ, vitamin A intake was estimated, which is representative of vitamin A intake spanning over the month before the semi-quantitative FFQ was administered. These patients were then contacted within a week of the administration of semi-quantitative FFQ, and a 24-hour recall was administered. From this 24-hour recall, the vitamin A intake was calculated. This was used as a measure with which the semi-quantitative FFQ was compared for validation.

Tools used in the study: The researchers employed two tools in this study

1. 24-hour recall method - This method involves assessing an individual's daily food intake by recording both the types of food and their quantities. To carry out the 24-hour recall, an interviewer asked each participant to recall their food consumption over

the previous 24 hours. The participants were queried about each meal in chronological order, beginning with the early morning, followed by breakfast, before lunch, and after breakfast, lunch, snacks, dinner, and food consumed just before sleeping. This 24-hour period started when they went to sleep on the initial day and ended at their bedtime the subsequent day. To aid in determining portion sizes, the researchers utilised Mohan's food atlas as a visual aid. Additionally, the participants were asked about any dietary supplements they were taking.

The 24-hour recall was administered to 55 pregnant women during the initial/first segment of the study who were present for their regular check-ups. In case any of the participants were not physically present to complete the semi-quantitative food frequency questionnaire (FFQ) or the 24-hour recall, their phone numbers were recorded so that the questionnaires could be administered to them separately.

For food items that required preparation, the researchers collected and verified recipes by preparing the foods in a metabolic kitchen. They then used the IFCT 2017 guidelines, along with Mohan's food atlas, to calculate the vitamin A content of the foods. Vitamin A intake was determined by multiplying the reported amount of each food item by the vitamin A values provided in the IFCT 2017 guidelines. The researchers calculated daily vitamin A intake from the 24-hour recalls using β-carotenoid values for vegetarian foods and retinol equivalents (REA) values for non-vegetarian foods and poultry products, as specified in the IFCT 2017 guidelines by the National Institute of Nutrition.

Vitamin A intake (RAE) was calculated by using the following formula:

$\mu g\ RAE = (\mu g\ retinol) + (\mu g\ \beta\text{-carotene}/12)$ [19]

2. Semi-quantitative Food Frequency Questionnaire (FFQ) - Food Frequency Questionnaires (FFQ) are typically qualitative measures of nutritional intake, providing rankings and comparisons. However, we have developed a semi-quantitative FFQ that includes specific measuring units for each type of food item tailored to the nature of the food. Pregnant women in the study were asked to recall their consumption of the listed food items over the past 30 days and were also questioned about the portion sizes they had consumed. To aid in portion size determination, participants were provided with visual assistance from Mohan's food atlas. The selection of food items for the semi-quantitative FFQ was conducted by compiling a list of items from 24-hour dietary recalls and subsequently narrowing down the list based on vitamin A content, following the guidelines outlined in the IFCT 2017. The final list was categorised into vegetables, fruits, staple food items, non-vegetarian and poultry, as well as additional supplements. Each semi-quantitative FFQ required approximately 15-20 minutes to administer, with an interviewer guiding the participants through the process. To ensure the reliability and validity of the FFQ, it was validated by comparing it to a 24-hour recall that was administered within a week of administering the semi-quantitative FFQ.

2.3. *Sample size*

A sample size of 100 is recommended for the validation of the semi-quantitative FFQ, as highlighted in previous review articles. [17,18]

2.4. *Statistical analysis*

The normality of the continuous data was tested using the Shapiro-Wilk test. Correlation analysis was done between 24-hour recall of vitamin A values and vitamin A values collected by semi-quantitative FFQ in group B for the purpose of semi-quantitative FFQ validation. The consistency of this agreement has been evaluated using Bland-Altman plots. Analyses were performed using SAS version 9.4 (SAS Institute, North Carolina, US). A p-value of ≤0.05 is considered statistically significant.

3. Results

A 24-hour food recall was administered to Group A (55 pregnant women), and information about their food intake was collected. From this recall data, we created a detailed list of 64 food items. After consulting the IFCT 2017 guidelines for nutrition released by the NIN and referring to the food atlas developed by the Madras Diabetes Research Foundation, we selected 38 food items with a relevant amount of vitamin A greater than 10ug/100gm REA in the form of β-carotenoid or retinol equivalents. These selected 38 food items were included in the semi-quantitative FFQ.

The normality of the continuous data was tested using the Shapiro-Wilk test (Table 2) test, which showed values above 0.05, indicating that the continuous data is normal in distribution without skew (Figure 4). Using the 24-hour recall method, the mean intake of vitamin A was found to be 523.51 ug REA with a standard deviation of 217.752 ug REA. The mean intake of vitamin A as measured via the semi-quantitative FFQ was found to be 870.347 ug REA with a standard deviation of 329.059 ug REA.

For validation of the questionnaire, 24-hour recall vitamin A intake was compared with the vitamin A intake measured by the Semi-quantitative FFQ, and the values were then checked for any correlation with Pearson and Spearman coefficient. The r value under the Pearson coefficient is 0.743 (95% CI = 0.613 - 0.834) with a p-value of <0.001, while the rho under the Spearman coefficient was 0.686 (95% CI = 0.535 - 0.794) with a p-value of <0.001

(Table 1) thus showing that there is significant correlation between 24-hour recall and Semi-quantitative FFQ vitamin A measurement.(Figure 1)

The consistency of this agreement has been evaluated using Bland-Altman plots. (Figure 5)

The study also showed that drumstick leaves, carrots, dates, and spinach are the major contributors of B carotene to the diet of the surveyed women as they consist of an average of 38%, 21%, 18%, and 12% to the total B-carotene intake in the diet.(Table 3)

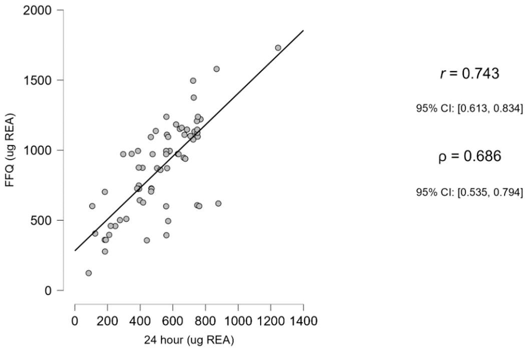

Figure 1: Scatter plot showing a significant correlation between data collected from 24-hour recall and data collected from FFQ in group B (r = 0.743, p<0.001 and rho =0.686, p<0.001)

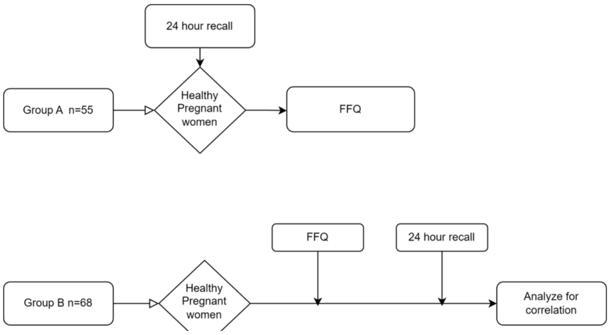

Figure 2: Representation of the tests administered to group A and group B

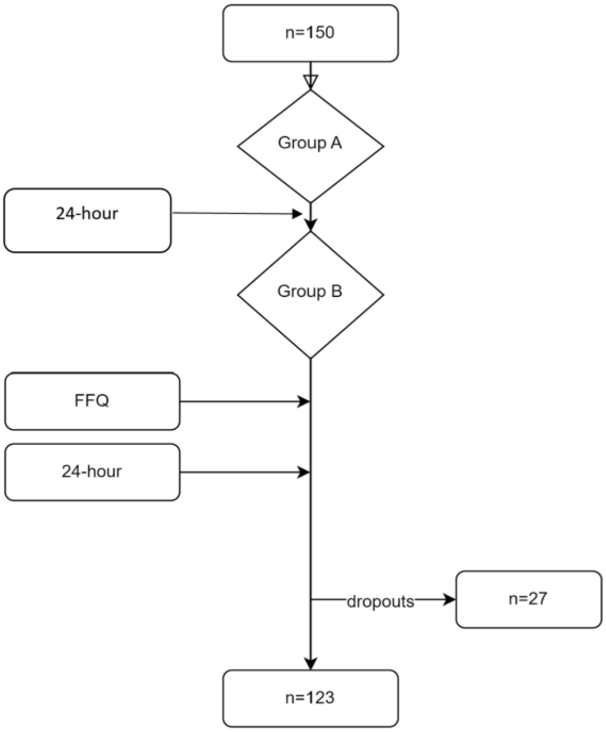

Figure 3: Flow diagram showing the temporal sequence of this study

4. Discussion

Pregnant women consist of a population that is at a particularly high risk of micronutrient deficiency. Due to the increased metabolic demands of pregnancy and the dire consequences of not satisfying them, maternal nutrition is a very important field of medicine.[20] Several micronutrients are integral for the well-being of both the mother and the fetus. The significance of folic acid lies in its ability to minimize the occurrence of neural tube defects in infants.[21] Iron becomes critical as it prevents anemia in mothers, a condition that can lead to issues like low birth weight and premature births.[22] The role of iodine is twofold it not only regulates thyroid functions but also plays a foundational role in a fetus' brain development.[23] Calcium is essential for the bone growth of the fetus and might also help in moderating blood pressure issues in pregnant women.[24] Vitamin D bolsters fetal bone formation and could also impact the fetus' immune responses.[25] Zinc contributes to the comprehensive development of the fetus and ensures maternal wellness.[26] Omega-3 fatty acids, especially EPA and DHA, are vital for the development of the baby's brain and vision.[27]

Vitamin A is one such important micronutrient with a multifaceted function during pregnancy. Vitamin A deficiency in pregnancy has been associated with iron deficiency anemia in the mother, which is particularly problematic in resource-limited settings.[28] This effect is not only seen in a state of deficiency but also in a state of relatively low intake of vitamin A via increased hepcidin synthesis in the liver.[29,30] Vitamin A is also vital for the ophthalmologic health of the mother, where a deficiency can lead to xerophthalmia. However, its effect has a wider reach, where lower vitamin A intake has been shown to be associated with higher maternal mortality, particularly within nutritionally deficient settings.[31] Vitamin A plays a critical role in the fetus's organ development, including the formation of the heart, lungs, kidneys, eyes, and bones, as well as the circulatory, respiratory, and central

Table 1: Showing a comparison between vitamin A values recorded by a 24-hour recall and FFQ of group B

Correlation Table						
	Pearson				Spearman	
	r		Lower 95% CI	Upper 95% CI	rho	
24 hour (ug REA) - FFQ (ug REA)	0.743	< .001	0.613	0.834	0.686	<.001

Table 2: Descriptive analysis of the data

Descriptive Statistics		
	FFQ (ug REA)	24 hour (ug REA)
Mean	870.347	523.511
Std. Deviation	329.059	217.752
Shapiro-Wilk	0.978	0.967
P-value of Shapiro-Wilk	0.268	0.072
Minimum	123.444	84.429
Maximum	1730.556	1244.571

Table 3: Average percentage of daily vitamin A provided

Food item	Percentage
Drumstick Leaves	38%
Carrot	21%
Date	18%
Spinach	12%

nervous systems.[32] On the other hand, excessive vitamin A intake, especially from supplements or medications like isotretinoin, can result in teratogenic effects leading to congenital disabilities such as abnormalities in the face, nervous system, and heart.[33] Given these potential dangers, pregnant women or those trying to conceive are advised to avoid excessive vitamin A sources and certain medications.[34] Striking the right balance of vitamin A intake is paramount for both maternal and fetal health, where a tool such as a validated FFQ would be preferable compared to expensive tests like serum retinol assays.

Food frequency questionnaires are an important tool for measuring dietary intake within epidemiological research.[35] There are three main components to FFQs - the list of foods, the frequency of consumption, and the portion sizes consumed. The frequency part of the questionnaire can be calculated using open-ended questions or predetermined frequency categories.[36] This allows us to inexpensively assess the nutritional status of the patient without the need for invasive investigations.

FFQa as a tool has been well studied, and several FFQs have been made and validated for pregnant women all around the world, but since India has a unique dietary pattern influenced by diverse cultures, traditions, and regions, it is crucial to have FFQs tailored specifically for each region and sub-population. Several previous attempts have been made to synthesize a FFQ for use in pregnant women of India,[37–39] but none so far have established a good correlation with vitamin A intake.

This paper discusses the creation and validation of a semi-quantitative FFQ, which can derive a measure of vitamin A intake over 30 days. Validation was achieved by comparing 24-hour recall data with data obtained from a food frequency questionnaire (FFQ). The study was conducted on a group of 123 patients that were divided into 2 groups - Group A, which consisted of 55 people, and Group B, which consisted of 68 people. This study was conducted in two phases where in the first phase, the FFQ specific to vitamin A was synthesized by creating a food list from group A. In the second phase, the FFQ was validated by comparing it to a 24-hour recall, which were both administered to the patients in group B.

The normality of the Vitamin A RAE values, that is, continuous data, was tested by the Shapiro-Wilk test, which showed values above 0.05, indicating that the continuous data is normal in distribution without skew.

To determine the relationship between the two methods of measuring vitamin A intake, correlation analysis with Pearson and Spearman coefficients was performed. To further calculate the reliability of this relationship, bland-altman plots were calculated.

The results showed a significant correlation between the 24-hour recall data and Semi-quantitative FFQ vitamin A intake. This was supported by the Pearson coefficient (r) of 0.743 with a p-value of less than 0.001, indicating a strong positive linear relationship between the two measures. Similarly, the spearman coefficient (rho) was 0.686 with a p-value of less than 0.001, reinforcing the

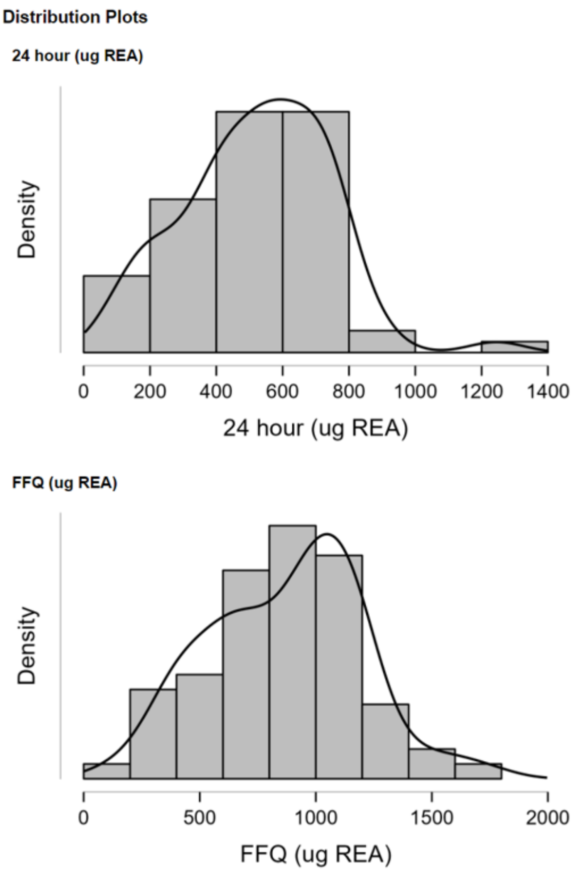

Figure 4: Density plot of the data that was collected

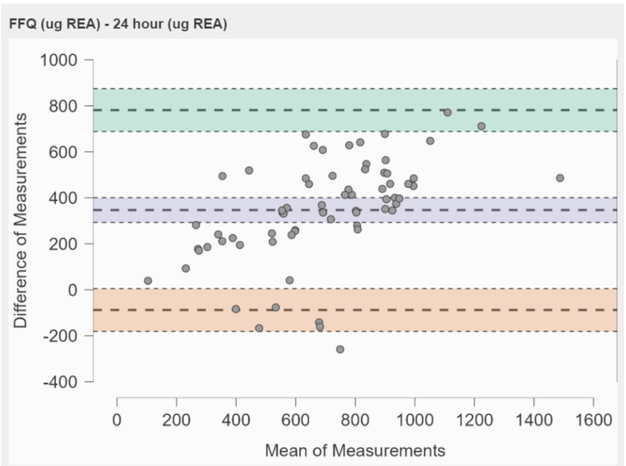

Figure 5: Bland Altman plot showing the consistency of the relationship between vitamin A measured by 24-hour recall and FFQ administered to group B

presence of a significant correlation. These results imply that the questionnaire used for assessing vitamin A intake is valid and can reliably estimate the intake based on the Semi-quantitative FFQ results. While the methodology we followed to get our results is the same as other studies, our findings are unique in that we are able to achieve a moderate-strong correlation that is significant between the Vitamin A values measured by the Semi-quantitative FFQ and 24-hour recall.

This may be because we have focussed solely on vitamin A, which all of the aforementioned studies did not do. While the studies did include Vitamin A, it was an inclusion in a larger set of micronutrients and macronutrients. Due to the overlapping nature of nutrition in food items where one item may be rich in multiple micronutrients, this could mean that food items that are relatively less rich in Vitamin A got excluded from the final food list as other food items took precedence and priority. This effect can be seen most prominently in one study in Brazil, where the correlation increased from 0.484 to 0.554 after the study focussed only on the food items that contain preformed Vitamin A.[40] Moreover, the south Indian population in Chennai, Tamil Nadu, that was studied has a relatively homogenous and constant diet pattern where the initial food list contained only 64 items compared to the 108 items long list used in a study conducted in Bangalore[38] and the 236 items long FFQ used in Pune.[39] This meant that the resulting FFQ, in our case, was a lot more concise and easier to administer, with less interview fatigue, which led to a higher degree of correlation. Furthermore, the paper presents information on the major contributors of vitamin A in the surveyed women's diet. Drumstick leaves, carrots, dates, and spinach were identified as the main sources of beta-carotene, accounting for an average of 38%, 21%, 18%, and 12% of the total intake, respectively. This means that these food items have the highest chance of working as a broad recommendation to prevent vitamin A deficiency in South Indian pregnant women living in Chennai.

Comparing these results with those of other research articles in the literature, it is important to note that validation studies of dietary measurement tools, such as FFQs, are commonly conducted to ensure their accuracy and reliability. Several studies have validated FFQs for assessing various micronutrients and macronutrients, utilizing different reference measures such as 24-hour recalls or diet records. This is the first study that shows a significant correlation with regard to vitamin A. This result shows that FFQs are more suitable and reliable if made for smaller, more homogenous communities and for specific nutrients rather than encompassing larger regions of populations and huge repertoires of nutrient measurements.

5. Conclusion

In conclusion, the results of this study suggest that the developed Semi-quantitative FFQ is a valid and useful tool for assessing vitamin A intake among pregnant women. The moderate-strong significant correlation between the Semi-quantitative FFQ and the 24-hour recall method emphasizes the utility of the Semi-quantitative FFQ in accurately estimating nutrient intake. Moreover, the identification of specific dietary contributors to β-carotene intake further strengthens the practical application of the Semi-quantitative FFQ in addressing nutritional deficiencies. However, it is important to note that the study's sample size was relatively small and limited to a specific group of patients, which may limit the generalizability of these findings. Future research could benefit from a larger sample size, and more research exploring specific micronutrients rather than a broader analysis would be extremely beneficial to the prenatal population.

6. Source of Funding

None.

7. Conflict of Interest

None.

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