



Development and validation of a food frequency questionnaire for assessing dietary vitamin D intake in young adults of Pakistan

Zainab Bohra ^{ID}¹, Sumaira Nasim ^{ID}¹, Sidra Zaheer ^{ID}¹, Habiba ^{ID}¹, Muhammad Zafar Iqbal Hydrie ^{ID}¹

ABSTRACT

Objectives: To develop and validate a culturally adapted food frequency questionnaire (FFQ) for assessing vitamin D (VD) intake among young Pakistani adults, using multiple 24-hour dietary recalls (24HDRs) as the reference method.

Methods: This cross-sectional study was conducted at Dow University of Health Sciences (DUHS) between August and October 2022. A VD-focused FFQ was developed using international food composition databases and adapted to Pakistani dietary practices, excluding culturally inappropriate or unavailable items. The final tool contained 53 items grouped into major food categories. Participants aged 18-30 years (n=99) were recruited through convenience sampling. Three interviewer-administered 24HDRs and two FFQs (FFQ1, FFQ2) were collected. Validity was assessed by comparing FFQ1 and FFQ2 with the mean 24HDRs using paired t-tests, Spearman's correlation, and Bland-Altman analysis, while reproducibility was evaluated by comparing FFQ1 and FFQ2.

Results: Mean VD intake was higher when estimated from FFQs (FFQ1: 5.96 μ g; FFQ2: 6.39 μ g) compared to 24HDRs (4.33 μ g), reflecting overestimation common to FFQs. Adequate VD intake (>2.5 μ g/day) was reported in >55% of participants across all methods. Spearman's correlation showed strong validity for FFQ1 vs. 24HDRs ($r = 0.693$, $p < 0.001$) and moderate validity for FFQ2 vs. 24HDRs ($r=0.538$, $p<0.001$). Reproducibility between FFQ1 and FFQ2 was strong ($r=0.700$, $p<0.001$). Bland-Altman indices were within acceptable ranges (3-5%), confirming agreement.

Conclusion: The culturally adapted VD-FFQ demonstrated moderate-to-strong validity and strong reproducibility, making it a reliable tool for estimating VD intake in young Pakistani adults. Its use may facilitate future research on dietary determinants of VD deficiency in this high-risk population.

Keywords: Nutritional Assessment (MeSH); Sports Nutritional Sciences (MeSH); Food Frequency Questionnaire (Non-MeSH); Vitamin D (Non-MeSH); 24-Hour Recall (Non-MeSH); Validation Study (MeSH); Pakistan (MeSH).

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INTRODUCTION

Hypovitaminosis D, or vitamin D deficiency (VDD), is a major global health concern affecting more than one billion people worldwide.¹ High prevalence has been documented in China, India, South America, and the Middle East.^{2,3} In Pakistan, VDD is particularly widespread across all age groups and genders, with 53.5% of the population affected. In Karachi alone, 76% of individuals have insufficient serum VD levels (<30 ng/mL), with a mean of 17.93 ng/mL.⁴ This is striking given the country's year-round sunlight exposure,⁵ yet deficiency remains

endemic due to cultural, lifestyle, and dietary factors.⁶ Limited outdoor activity, modest clothing, indoor living, poor housing, and specific dietary practices restrict sunlight exposure and VD synthesis. Moreover, low nutritional awareness, suboptimal cooking methods, and habits such as betel nut chewing further impair VD metabolism.⁷

Although VD was initially recognized for its role in bone health and calcium absorption, growing evidence highlights its importance in numerous physiological processes within the body.⁸ Its deficiency is now linked not only to rickets and osteoporosis but also

I: School of Public Health, Dow University of Health Sciences, Karachi, Pakistan

Email ✉: sumaira.nasim@duhs.edu.pk

Contact #: +92-300-8379692

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to diabetes, cardiovascular diseases, tuberculosis, autoimmune disorders (e.g., psoriasis, multiple sclerosis), and cancers of the breast, prostate, and colon.^{9,10}

While sunlight is the primary natural source of vitamin D, dietary intake also contributes significantly.¹¹ Adequate intake through food is essential, as demonstrated by Crowe FL, et al., who reported higher serum vitamin D levels among meat and fish eaters compared to vegetarians and vegans.¹¹ Despite this, fulfilling daily requirements remains a challenge. According to the Pakistan Dietary Guidelines for Better Nutrition (PDGN), the recommended dietary allowance (RDA) is 2.5 μ g for adults and 10 μ g for pregnant and lactating women.¹⁰ In contrast, international guidelines advise higher intakes, recommending 15 μ g (600 IU) for adults and 20 μ g (800 IU) for individuals over 70 years.

Assessing vitamin D intake poses considerable challenges, underscoring the need for reliable dietary assessment tools. Although serum 25(OH)D concentration remains the gold standard for evaluating vitamin D status, it is invasive and costly. In contrast, the Food Frequency Questionnaire (FFQ) is widely used in nutritional research due to its affordability, non-invasive nature, and ease of administration.¹³⁻¹⁵ Improving the accuracy of dietary intake estimates is essential for understanding diet-health associations. This can be achieved by employing modern technologies for dietary reporting, integrating self-reported data with biomarkers, and combining multiple self-report instruments.¹³⁻¹⁵ The 24-hour dietary recall (24HDR) is

commonly employed as a reference method in FFQ validation studies.¹³ Previous research by Taylor C, et al.,¹⁵ Pitchard et al.,¹⁶ and Głabbska JM, et al.,¹⁷ has validated different FFQs for assessing vitamin D intake across diverse populations. While many epidemiological studies adapt existing FFQs, wide dietary variations highlight the need for culture-specific tools. In Pakistan, distinct foods, serving sizes, and cooking practices necessitate a tailored FFQ that accurately reflects local dietary habits.⁴ Despite the high prevalence of vitamin D deficiency, dietary assessment of vitamin D intake remains underexplored. Therefore, this study aimed to develop and validate a culturally specific, vitamin D-focused FFQ against the 24HDR in young Pakistani adults.

METHODS

Recruitment of participants: This cross-sectional study was designed to develop and validate culture-specific vitamin D (VD) FFQ. Participants aged 18–30 years were recruited through non-probability (convenience) sampling, using social media platforms, student clubs, and email invitations within Dow University of Health Sciences (DUHS). Eligible participants were DUHS students residing in Pakistan. Exclusion criteria included enrollment in nutrition programs, adherence to special diets, pregnancy or lactation, and the presence of diet-related chronic diseases. Both the 24HDR and the FFQ were administered through interviewer-led sessions. Participants were informed about the study objectives and were notified that they would be contacted at least five times for data collection.

Given the use of convenience sampling, the results may not be fully generalizable to all Pakistani adults; however, they provide valuable insights into this specific population.

Ethical consideration: Ethical considerations were taken in this study, including obtaining informed consent, ensuring privacy and confidentiality, and avoiding any potential harm or exploitation of participants. The research protocol has been approved by the review board of Dow University of Health Sciences and was registered as

IRB-1121, approved on 1st October 2022.

Training of volunteers: Data collection was carried out by a team of trained volunteer interviewers. Prior to the study, they completed a two-day training program covering essential topics, including research ethics and dietary recall techniques, with particular emphasis on the multiple-pass approach.

Piloting of questionnaire: Prior to data collection, the preliminary version of the VD-FFQ was pilot-tested on a sample of 10 individuals who met the inclusion criteria but were not part of the main study. Revisions were made based on the pilot study findings to improve the tool's validity, reliability, and clarity, and to address any issues identified.

Validation of VD FFQ: This validation study was conducted over a three-month period (August–October 2022). The VD-FFQ was evaluated for both validity and reproducibility, following the framework described by Willett WC, et al.,¹⁸ Validity (external validation) was assessed by comparing FFQ results with 24HDRs, while reproducibility (internal validation) was examined by comparing results from two administrations of the FFQ (FFQ1 and FFQ2). All assessments were carried out by the same researcher.

Data Collection procedure

24-hours dietary recalls: In the first stage, three 24-hour dietary recalls (24HDRs) were collected from each participant by trained volunteers, covering the full day from morning to night. Interviewers probed for any missed foods and recorded the time and location of each meal. Detailed information on portion size, condiments, preparation method, and brand was documented. To improve accuracy, household utensils, standard measuring tools, and both 2D and 3D food models were used as visual aids. To account for variation in eating patterns, two weekdays and one weekend day were randomly selected per participant. Data were analyzed using the CRON-O-METER application to estimate average daily vitamin D intake ($\mu\text{g/day}$).

VD dietary intake food frequency questionnaire: The FFQ was developed to estimate vitamin D intake, focusing solely on food items that serve as dietary sources of vitamin D. Frequently consumed items with a vitamin D content of $\geq 0.01 \mu\text{g}$ per serving were selected and incorporated, drawing on previously validated vitamin D FFQs.¹⁹ Since the Pakistani food composition table does not provide data on vitamin D content, the USDA Food Composition Database was used as the primary reference. Where USDA data were unavailable, additional information was obtained from the Nutrition Coordinating Center (NCC), the National Institutes of Health (NIH), the UK vitamin D FFQ,²⁰ and the Polish vitamin D FFQ.²¹ Based on these sources, a final 53-item FFQ was constructed.

During cultural adaptation, haram (forbidden) foods and those uncommon or unavailable in Pakistan were excluded. The questionnaire items were categorized into five main food groups: milk and milk products, cereal grains and grain products, vegetables, desserts/sweets and miscellaneous, and butter and spreads. Meat, pulses, and eggs were grouped together with other protein-rich foods. Serving sizes were confirmed during the pilot study. Participants were asked to report their usual consumption frequency for each item according to their dietary habits. Additional questions addressed vitamin D supplement use, including brand, dosage, and frequency. Both the 24HDRs and the FFQ relied on self-reported dietary intake.

Computerized format: The VD FFQ was designed as part of an electronic survey (e-survey) using Google Forms. Instructions and examples for completing the e-FFQ were included. The e-survey was designed to ensure all questions were answered before submission, preventing missing data. Food items were listed under their respective food groups. The FFQ's frequency of intake section had nine categories: never/less than once a month, monthly, once a week, twice a week, three times a week, four to five times a week, twice a day, twice or thrice a day, and four to five times a day.

Separate frequency options for fats, oils, and spreads were provided, including specific measurements such as 1 tea spoon (tsp), 1 table spoon (tbsp), 2 tbsp, 3 tbsp, ½ cup, 1 cup, 2 cups, or none. Results were collected upon completion and sorted on a Google Form spreadsheet. Results were collected via Google Sheets. Participants who completed FFQ1 were reminded via email/WhatsApp to complete FFQ2. Each form took ~15 minutes. Online forms reduced completion time, ensured all responses were recorded, and minimized fatigue compared to paper versions.

Sample size calculation: The sample size was calculated using OpenEpi. The prevalence of vitamin D deficiency among young adults was assumed to be 53.0%. A correlation coefficient of 0.540 between the FFQ and three 24HDRs for daily vitamin D intake was used, with 80% power and a 95% confidence interval. This yielded a required sample size of 88 students. To account for potential non-response and missing data, a 15% attrition rate was added, increasing the target to 101 participants. Of the 122 students initially recruited, 99 completed all study requirements and were included in the final analysis.

Data analysis

Validation of FFQ: According to Willett WC, et al,¹⁸ the evaluation of the VD-FFQ involves assessing both reliability and validity. Reliability (internal validation) was examined by comparing the results of two administrations of the FFQ (FFQ1 and FFQ2). Validity (external validation) was assessed by comparing VD intakes estimated from the FFQ with those obtained from three 24HDRs. Statistical analyses included paired t-tests, Spearman's correlation coefficients, and Bland-Altman plots.

Analysis of 24hrs dietary recalls (Reference method): All 24HDRs were analyzed using CRON-O-Meter, a web-based nutrient analysis tool that tracks biometric and dietary data, providing information on more than 60 nutrients across a database of over 7,500 food items. This software was selected for its comprehensive

international food database. For analysis, the food items most closely matching the reported foods were selected. Serving sizes were determined using available menu information; when not specified, standard unit values (e.g., one whole fruit for "grapefruit" or one cup for "orange juice") were applied. In the case of mixed dishes with similar recipes, one serving was counted for each item unless specific menu details indicated otherwise.

Analysis of VD content from the FFQ: For items reported with weekly or monthly frequencies, the total servings were divided by seven or thirty to obtain a daily serving estimate. Vitamin D intake from each food item was calculated using the following formula:

VD intake (μg) = daily number of servings \times vitamin D content per serving.

The average daily dietary vitamin D intake (μg) was then determined by summing the intake values across all food groups.

Statistical analysis: All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS), version 21.0.0 (SPSS Inc., Chicago, IL, USA). Descriptive statistics (mean, standard deviation, and percentages) were calculated to summarize participant characteristics. Categorical variables were presented as frequencies and percentages.

For validity analyses (FFQ1 vs. 24HDR and FFQ2 vs. 24HDR) and

reproducibility analyses (FFQ1 vs. FFQ2), data distribution was assessed using the Shapiro-Wilk test. Given the nonparametric distribution, Spearman's rank correlation was applied. Mean of three 24HDRs was used for all analyses. Results were compared against the recommended dietary allowance (2.5 $\mu\text{g/day}$ of vitamin D) specified by the Pakistan Dietary Guidelines for Better Nutrition (PDGN).

Bland-Altman plots were constructed to evaluate agreement in both validity (FFQ1 vs. 24HDR and FFQ2 vs. 24HDR) and reproducibility (FFQ1 vs. FFQ2). The Bland-Altman index was interpreted, and limits of agreement (LOA) were calculated as the mean difference in vitamin D intake between the two methods \pm 1.96 times the standard deviation of the differences. Outliers were identified, and sensitivity analyses were performed by excluding extreme intake values to confirm the robustness of the findings.

RESULTS

Descriptive characteristics: Participants were aged 18-30 years, with a mean age of 21.38 ± 1.88 years. The overall questionnaire response rate was 80% (99 of 122). Females comprised 65% of the sample, while 34% were males, and the majority were undergraduate students (90%).

The mean vitamin D intake estimated from the FFQs was consistently higher (5.96 μg for FFQ1 and 6.38 μg for FFQ2) compared to that from the three 24HDRs (4.33 μg). This overestimation is consistent with findings from other

Table 1: Comparison of vitamin D intake estimated by 24-hour dietary recalls and food frequency questionnaires (FFQ1, FFQ 2)

Category		Three 24hrs diet recalls	FFQ1	FFQ2
Mean \pm SD (μg) ²		4.33 \pm 2.74	5.96 \pm 4.25	6.39 \pm 4.49
Median (μg)		3.47	5.46	6.15
Minimum (μg)		0.70	0.01	0.07
Maximum		11.33	17.30	17.02
Percentage of individuals (characterized by intake of 2.5 μg)	Adequate intake (%)	68.7%	77.7%	76.8%
	Inadequate intake (%)	31.3%	28.3%	23.2%

FFQ: Food frequency questionnaire

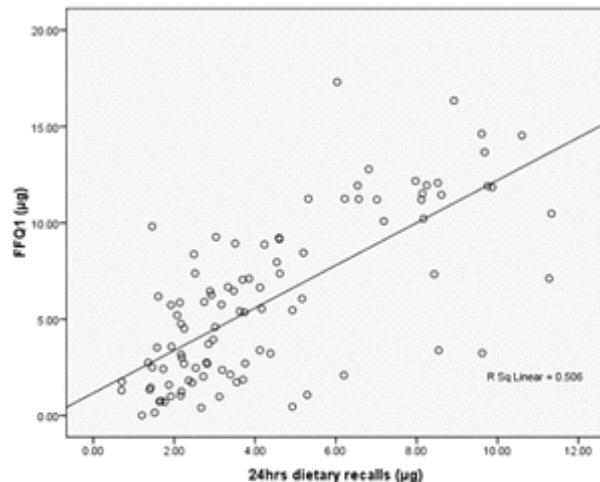


Figure 1: Analysis of correlation between VD FFQ1 and the three 24hrs dietary recalls for VD daily intake.

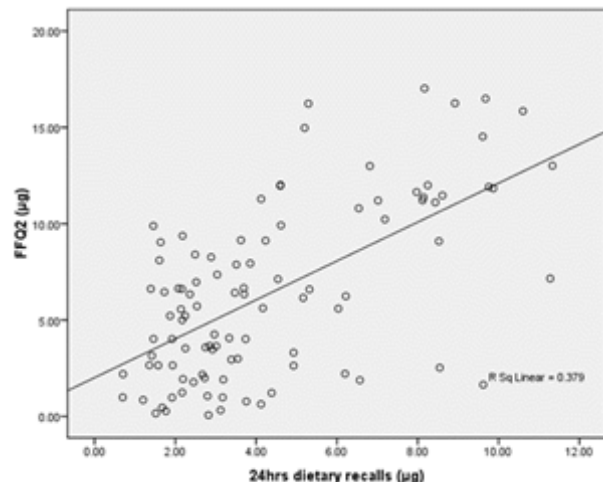


Figure 2: Analysis of correlation between VD FFQ2 and the three 24hrs dietary recalls for VD daily.

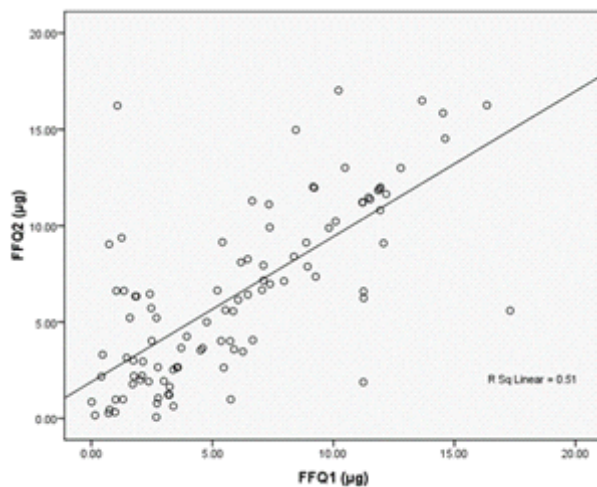


Figure 3: Analysis of correlation between VD FFQ1 and FFQ2 of VD daily intake.

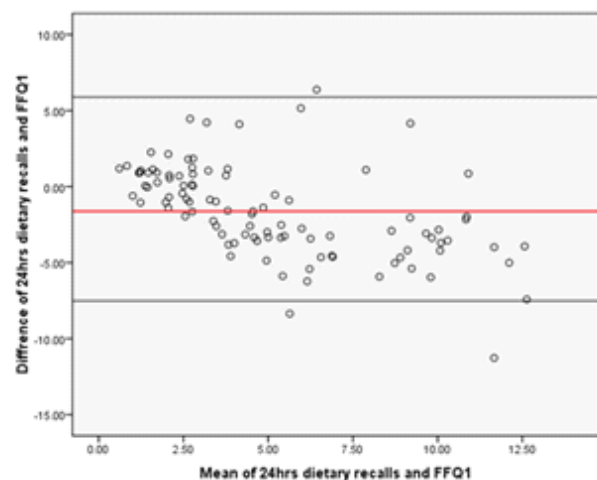


Figure 4: Bland-Altman plot comparing VD-FFQ1 with the three 24hrs DR for VD daily intake (Bland-Altman index of 3%).

FFQ validation studies and may be attributed to recall bias, reduced day-to-day variability captured by the FFQ, and portion size overestimation.

Nevertheless, the proportion of participants meeting the recommended intake ($>2.5 \mu\text{g/day}$, as per Pakistani Dietary Guidelines¹²) remained high across methods, with $>55\%$ classified as adequate. Fewer than 7% of participants reported using vitamin D supplements.

Vitamin D intake estimated by FFQs was consistently higher than that from 24HDRs. Median intake was $3.47 \mu\text{g}$ with 24HDRs compared to $5.46 \mu\text{g}$ and $6.15 \mu\text{g}$ with FFQ1 and FFQ2, respectively. Overall, more than two-

thirds of participants met the adequacy threshold ($>2.5 \mu\text{g/day}$) across all methods, with slightly higher adequacy rates reported by the FFQs (Table 1). Compared with the recommended level of $2.5 \mu\text{g}$,¹³ dietary intake was adequate in more than half of the participants. Intake estimated by the 24HDR was lower than that obtained from the VD-FFQs, while results from FFQ1 and FFQ2 were largely comparable.

Analysis of correlation: Figure 1 shows the correlation between FFQ1 and the three 24HDRs for daily vitamin D intake, with a Spearman coefficient of $R=0.693$ (95% CI: 0.55–0.79, $p<0.0001$), indicating a statistically

significant strong positive association.

The analysis of the correlation between FFQ2 and the three 24hrs-DRs for VD daily intake is presented in Figure 2. The Spearman rank correlation coefficient $R=0.538$ (95% CI: 0.37–0.67), $p<0.0001$, reveals highly significant but moderate association for VD daily intakes obtained using a validated method of VD (FFQ2) and the three 24hrs-DRs.

The analysis of correlation between FFQ1 and FFQ2 of VD daily intake is presented in Figure 3. The Spearman rank correlation coefficient was applied where $R=0.700$ (95% CI: 0.57–0.80), $p<0.0001$, it revealed statistically

significant strong association for VD daily intakes obtained using a VD FFQ between FFQ1 and FFQ2.

These findings suggest that while FFQs show moderate-to-strong validity against 24HDRs, the agreement is stronger when assessing reproducibility between FFQ1 and FFQ2.

Bland altman plots: The Bland-Altman plot was constructed to assess the strength of agreement between VD dietary intake data obtained from three 24hrs-DRs and VD FFQ1. In Figure 4, the mean difference was $-1.623 \mu\text{g}$, with limits of agreement (LOA) ranging from -7.512 to $4.266 \mu\text{g}$. Three participants (3%) fell outside the LOA, yielding a Bland-Altman index of 3%. This indicates acceptable agreement, although FFQ1 systematically overestimated intake relative to 24HDRs.

In Figure 5, the Bland-Altman plot comparing FFQ2 with the three 24hrs DRs for VD daily intake is presented. The mean difference was $-2.053 \mu\text{g}$, with LOA from -9.001 to $4.895 \mu\text{g}$. Four participants (4%) were outside the LOA, corresponding to a Bland-Altman index of 4% was obtained for the LOA after adding a ± 1.96 -fold standard deviation.

Again, FFQ2 tended to overestimate intake compared to 24HDRs, though within acceptable error margins. The Bland-Altman plot comparing FFQ2 with FFQ1 of VD daily intake is presented in Figure 6. The mean difference was $0.429 \mu\text{g}$, with LOA from -6.074 to $6.932 \mu\text{g}$. Five participants (5%) were outside the LOA (Bland-Altman index=5%). These results demonstrate that reproducibility between FFQs was stronger than validity against 24HDRs, consistent with expectations for self-administered dietary tools.

DISCUSSION

This study outlines the development and validation of a vitamin D-specific FFQ for Pakistani adults aged 18-30 years, representing the first culturally tailored tool for this purpose. The FFQ estimated to be significantly higher intakes than 24HDRs, though the two methods showed moderate-to-strong

correlations. Overestimation by the FFQ likely reflects recall bias, portion size misclassification, and limited day-to-day variability, whereas 24HDRs may underestimate intake due to reliance on short-term memory and non-representative recall days. These findings emphasize the complementary limitations of both methods, with FFQs tending to overestimate and 24HDRs to underestimate vitamin D intake.

Several investigators have demonstrated that FFQs are useful for assessing vitamin D intake in population studies.^{20,22} For instance, Kiely M, et al.,¹³ validated a VD-specific FFQ against diet history and found higher intakes with a strong correlation ($R=0.71$, $p<0.001$). Weir RR, et al.,¹⁴ also validated a 17-item FFQ against a 4-day weighed food record, showing higher VD intake estimates from the FFQ with strong agreement between the two methods.

Several investigators showed that FFQ can be used for the assessment of VD in population studies.^{20,22} For example, a study validated an FFQ to assess VD intake among young women (20–30 years old) in Croatia, reporting higher intakes of VD with FFQ compared to dietary records (132 IU vs. 76 IU). Kiely M, et al.,¹³ validated a VD-specific FFQ against diet history and found higher intakes with FFQ, with a strong association ($R=0.71$, $p<0.001$). Similarly, Weir et al.¹⁴ validated a 17-item FFQ against a 4-day weighed food record. The results should be a higher intake of VD from the FFQ as compared to the food records with a strong correlation between these two assessment methods.

In dietary validation studies, correlations exceeding 0.7 are rare due to the inherent complexity of dietary assessment.¹⁵ Thus, the coefficients observed in this study ($R=0.693$ for FFQ1 vs. 24HDRs and $R=0.538$ for FFQ2 vs. 24HDRs) should be regarded as satisfactory rather than strictly “strong.” Importantly, reproducibility between FFQ1 and FFQ2 ($R=0.70$) was higher than validity, aligning with findings from other dietary validation research.

The level of agreement between the FFQ and 24HDRs for vitamin D intake

was evaluated using Bland-Altman (BA) plots, a widely accepted method and often considered the “gold standard” for validation.^{19,29} The BA index was 3% for FFQ1 vs. 24HDRs, 4% for FFQ2 vs. 24HDRs, and 5% for FFQ1 vs. FFQ2. These values are comparable to previous reports, such as Prichard JM, et al.,¹⁶ (6.7%) and Taylor Taylor C, et al.,¹⁵ (5.3%). Thus, the agreement observed in this study is consistent with published literature and indicates that the FFQ achieved satisfactory validity. Importantly, the BA indices were all below the 10% threshold set a priori for positive validation, suggesting that although FFQs tended to overestimate intake, the degree of misclassification remained within acceptable limits.²¹

Accurate assessment of VD intake is difficult due to high day-to-day variance in consumption of VD rich foods, particularly fish.²³ However, because fish products, if consumed, contribute to the intake of VD they may be the primary source of errors while the intake is assessed based on the dietary recall or record. In a Swedish study, the highest VD intake was observed in those who consumed the highest number of fish.²⁴

Assessing adult food intake is challenging due to underreporting, limited dietary diversity, participant, and research error, and selecting an assessment method. Accurate assessment is essential, but long-term assessments are impractical for epidemiological research due to the increased burden on participants and the time needed to evaluate the data.

A number of VD specific FFQ targeting South Asian population were developed in Canada^{25,26} and United Kingdom,^{20,27} all these studies were conducted on South Asian migrant residing. The matter of inadequate VD levels carries significant weight as a pressing health concern within the South Asian population.^{4,16,28}

In Pakistan, studies have been conducted to evaluate VD status and its relationship with diseases.²⁹ No study has employed FFQ to assess VD dietary intake.²⁸ The VD FFQ in this study is based on foods and beverages commonly consumed in Pakistan. The present tool is based on Pakistani dietary habits and may be adapted for

fortified foods, but its reliance on foreign (USDA) food composition tables introduces potential inaccuracies. Development of locally validated databases would improve precision.

Our findings revealed that over 55% of the population consumes $\geq 2.5 \mu\text{g}$ of vitamin D daily, in accordance with the current Pakistani Dietary Guidelines, which suggest a daily allowance of $2.5 \mu\text{g}$. However, international standards, such as those from the National Research Council, Institute of Medicine, Washington, DC, and the National Institutes of Health (NIH) Office of Dietary Supplements (ODS), recommend a dietary allowance of 600 IU (equivalent to $15 \mu\text{g}$)^{14,15} and India recommends $10 \mu\text{g}$ (400 IU) under low sunlight.³⁰ The current RDA for vitamin D in Pakistan, at $2.5 \mu\text{g}$, appears notably low given widespread deficiency and limited sun exposure due to cultural clothing practices. Thus, local guidelines may need to be revised upward in light of both international benchmarks and the deficiency burden.

This study validated the FFQ using 24HDRs, acknowledging that no dietary reference method is perfectly accurate. Although biomarker analysis offers an independent approach, it was not applied here for vitamin D estimation. Research suggests validating FFQs by comparing nutritional biomarkers with another dietary assessment method to ensure accuracy. Serum 25(OH)D concentration is generally a reliable indicator of vitamin D status and is often considered in validation studies; however, its use is complex for vitamin D, as sunlight exposure substantially influences serum levels, unlike most other nutrients.

Biomarker validation using serum 25(OH)D was not performed in this study, which limits comparability with international data. However, sunlight exposure makes serum 25(OH)D an imperfect dietary validation marker for vitamin D. Previous studies developing vitamin D-specific FFQs have similarly omitted biomarker analysis, as it is invasive, costly, and burdensome for participants.^{18,19} Biomarkers are best used alongside other dietary assessment methods rather than as stand-alone references. FFQs are also prone to errors related to portion size

estimation, recall bias, and incomplete food lists.¹⁷ Therefore, beyond their development and initial validation, FFQs require adaptation and revalidation across diverse populations. Tools tailored to specific cultural or geographic contexts are essential to ensure accuracy and applicability.²⁷

Limitations of the study

This study has several limitations. First, the use of convenience sampling within a single university and a restricted age range (18–30 years) limits the generalizability of findings to the broader Pakistani population. Second, reliance on three 24-hour dietary recalls may not have fully captured intra-individual variation in vitamin D intake, which is often irregular. Third, data collection was conducted only between August and October, overlooking seasonal variation in sunlight exposure and the availability of vitamin D-rich foods such as fish. Fourth, intake estimation relied on foreign food composition databases (USDA, UK, Polish, NCC) in the absence of locally validated data, which may have introduced inaccuracies. Finally, as with all self-reported tools, recall bias and portion size estimation errors remain inherent challenges.

Despite these limitations, this study provides an important first step in developing a culturally specific vitamin D FFQ for Pakistan, offering a foundation for future validation across diverse populations, seasons, and with biomarker support.

CONCLUSION

This study developed and validated the first culturally adapted vitamin D-specific FFQ for young adults in Pakistan. In the absence of locally available vitamin D data within national food composition tables, this tool provides a practical option for dietary assessment. The FFQ demonstrated satisfactory validity against 24-hour dietary recalls and strong reproducibility between repeated administrations, although it tended to overestimate intake relative to recalls. Despite these limitations, agreement remained within acceptable ranges and consistent with international validation studies. Future research should extend

validation across other age groups, geographic settings, and seasons, ideally incorporating biomarker support, to strengthen its applicability and generalizability.

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AUTHORS' CONTRIBUTION

The Following authors have made substantial contributions to the manuscript as under:

ZB: Conception and study design, acquisition, analysis and interpretation of data, drafting the manuscript, approval of the final version to be published

SN & MZIH: Conception and study design, critical review, approval of the final version to be published

SZ: Analysis and interpretation of data, drafting the manuscript, approval of the final version to be published

Ha: Conception and study design, acquisition of data, critical review, approval of the final version to be published

Authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

CONFLICT OF INTEREST

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The data that support the findings of this study are available from the corresponding author upon reasonable request



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KMUJ web address: www.kmuj.kmu.edu.pk

Email address: kmuj@kmu.edu.pk