

# Development and Validity of a Food Frequency Questionnaire for Assessing Vitamin C Intake in Turkish Adults

## Yetişkin Türklerde C Vitamini Alımının Değerlendirilmesine Yönelik Besin Tüketim Sıklığı Anketinin Geliştirilmesi ve Geçerliliği

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**ABSTRACT Objective:** This study aimed to evaluate the validity of a food frequency questionnaire (FFQ) developed to determine vitamin C intake. **Material and Methods:** This research was carried out at İstanbul University Faculty of Dentistry between October 2017 and May 2018. There were 75 individuals in this study, who were 18-65 years old. In order to determine the level of vitamin C intake of individuals, a FFQ was formed which contains the consumption frequency and the amount of consumption of 83 foods and its language is Turkish. After the determination of the plasma ascorbic acid level (PAAL) of the individuals, the validity of the questionnaire was examined by Spearman correlation test, Bland Altman plot and kappa statistics. All statistical analysis were performed using the IBM SPSS Statistics 20 Software and statistically significant differences were determined using a p value <0.05. **Results:** A total of 75 people (31 male and 44 female) participated in this study, and the mean age was 35.69±10.35 years. The median PAAL was 7.94 mg/L, while the median intake of vitamin C was 136.84 mg/d. There was a positive correlation between PAAL and intake of vitamin C (r=0.41; p<0.001). The values of PAAL and intake of vitamin C calculated by FFQ were divided into quartiles, and the rate of the classifying into the same or adjacent quartile was 81.4%. **Conclusion:** In conclusion, this questionnaire can be used to determine intake of vitamin C in Turkish adults.

**Keywords:** Ascorbic acid; validation studies; nutritional status

**ÖZET Amaç:** Bu çalışma, C vitamini alımını belirlemek amacıyla geliştirilen bir besin tüketim sıklığı anketinin (food frequency questionnaire: FFQ) geçerliliğini değerlendirmeyi amaçlamaktadır. **Gereç ve Yöntemler:** Bu çalışma, Ekim 2017 - Mayıs 2018 tarihleri arasında İstanbul Üniversitesi Diş Hekimliği Fakültesi'nde gerçekleştirilmiştir. Çalışmaya 18-65 yaş arasındaki 75 gönüllü birey katılmıştır. Bireylerin C vitamini alımlarını belirlemek için, toplam 83 besinin tüketim miktarı ve sıklığını içeren Türkçe bir besin tüketim sıklığı anketi oluşturulmuştur. Bireylerin plazma askorbik asit seviyelerinin belirlenmesinin ardından anketin geçerliliği Spearman korelasyon testi, Bland Altman grafiği ve kappa istatistiği ile incelenmiştir. Tüm istatistiksel analizler IBM SPSS Statistics 20 yazılımı kullanılarak yapılmış ve p<0,05 değeri istatistiksel olarak anlamlı kabul edilmiştir. **Bulgular:** Çalışmaya katılan 75 kişiden 31'i (%41,3) erkek, 44'ü (%58,7) kadındır. Medyan plazma askorbik asit seviyesi 7,94 mg/L iken, medyan C vitamini alımı 136,84 mg/gün'dür. Plazma askorbik asit seviyesi ile C vitamini alımı arasında pozitif bir korelasyon bulunmuştur (r=0,41; p<0,001). Plazma askorbik asit düzeyleri ve FFQ ile hesaplanan C vitamini alım miktarları çeyreklerle ayrılmış; aynı veya bitişik çeyrekte sınıflandırma oranı %81,4 olarak bulunmuştur. **Sonuç:** Sonuç olarak, bu anket yetişkin Türklerde C vitamini alım düzeyini belirlemek için kullanılabilir.

**Anahtar Kelimeler:** Askorbik asit; doğrulama çalışmaları; beslenme durumu

There are many methods used in the evaluation of food consumption in epidemiological studies. The most frequently used method in this studies are 24-hour dietary recall and food frequency questionnaire

(FFQ). The FFQ is often used in studies examining nutrient-disease interactions, while 24-hour dietary recall is used to determine groups that may need nutritional guidance. Some errors may occur in both of

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these methods. People tended to forget (especially for FFQ), the consumption of certain foods may be reported incorrectly, or mistakes in the calculation of energy and nutrient values of foods may occur.<sup>1-5</sup> Various FFQs were conducted to evaluate the intakes of certain nutrients, but their performance needs to be assessed before they can be used. The performance of a FFQ can be determined by comparing the results of various methods, comparing the biochemical indicators, or validity / reliability tests.<sup>6</sup>

Vitamin C is one of the water-soluble vitamins and it is synthesized from glucose in the liver of most mammals. Humans need to take vitamin C with foods, because they do not have L-gulonolactone oxidase enzyme.<sup>7,8</sup> The most important sources of this vitamin are citrus, strawberry, kiwi, broccoli, spinach, pepper, rosehip, parsley. The content of vitamin C of foods varies according to harvesting time, storage time and conditions, preparation and cooking methods.<sup>9-11</sup> Dietary intake of vitamin C can be measured in two ways: assessment of nutritional status (FFQ, dietary record and 24-hours dietary recall) or measurements of serum plasma vitamin C levels.<sup>12</sup> While assessing the validity of a FFQ aiming to determine the amount of vitamin C intake; age, gender, smoking habits, infections and other factors should be considered. In addition, since vitamin C is not stable, the sample should be carefully preserved. Plasma ascorbic acid level (PAAL) is a good biomarker of intake of about 30-90 mg/day of vitamin C.<sup>13</sup> The reference value for PAAL is 0.5-1.8 mg/dL; below 0.3 mg/dL is 'inadequate' and above 3.0 mg/dL is 'over'.<sup>14</sup> Storage capacity of vitamin C in the body is 1500 mg, if this storage is less than 350 mg, deficiency may occur. The prevalence of deficiency in the world varies such as 7.1% in the USA and 73.9% in North India. Eight-12 weeks of inadequate intake leads to the appearance of symptoms.<sup>15</sup> Symptoms of vitamin C deficiency in early period are fatigue, loss of appetite, delay in wound healing; symptoms in the long-term are anemia, susceptibility to infections, tooth loss, gingival bleeding and growth retardation.<sup>16</sup>

It is important to determine vitamin C deficiency in the community. The aim of this study is to evaluate the validity of a FFQ developed to determine the intake of vitamin C.

## MATERIAL AND METHODS

### STUDY DESIGN

This research was carried out at Istanbul University Faculty of Dentistry between October 2017 and May 2018. This study included the relatives of 75 patients who came to the institution, and between 18-65 years, who were not using alcohol or cigarettes, were neither pregnant nor breast-feeding mothers, and did not have any systemic or oral disease.

This study is part of the project entitled 'The relationship between vitamin C and oxidative status in adults with chronic periodontitis' approved by the Marmara University School of Medicine Clinical Research Ethics Committee (Protocol no: 09.2017.035). The sample group of this study constituted the control group of the project.

All procedures were carried out in accordance with the Declaration of Helsinki. All individuals were informed about this study, then informed consent forms were obtained.

### DEVELOPMENT OF THE FFQ AND ASSESSMENT OF DIETARY INTAKE OF VITAMIN C

While developing the questionnaire, as a first step, the foods that are the source of vitamin C were identified. Vitamin C contents of foods were obtained from BeBiS Nutrition Information System 7.1 (Pacific Company, Istanbul, Turkey), United States Department of Agriculture (USDA) Food Composition Databases and Turkish food composition database (TURKOMP); and then foods containing more than 2 grams of vitamin C in 100 grams were listed. The foods that are inaccessible in Turkey and the foods that are not suitable for Turkish food culture were removed from the list. In the pilot study, this FFQ was applied to 45 adults living in different districts of Istanbul, and the foods that were never consumed in the last year were removed from the list.

The 83 foods included in the FFQ were categorized under 3 headings as *Vegetables*, *Fruits* and *Drinks and other foods*. In this questionnaire, the consumption frequency (every meal, every day, 5-6 times a week, 3-4 times a week, 1-2 times a week, once in 15 days, once in a month, never) and consumption

amount of 83 foods containing vitamin C in the last month were questioned. It is also noted that these foods were generally consumed as raw or cooked, due to the loss of vitamin C during cooking.

This questionnaire was applied to 75 individuals face to face. The visual equipment was used in the process of determining the amount of the foods.<sup>17-18</sup> Vitamin C contents of foods were obtained from the BeBiS Nutrition Information System 7.1 (Pasifik Company, Istanbul, Turkey), United States Department of Agriculture (USDA) Food Composition Databases and Turkish food composition database (TURKOMP). In addition, the consumption frequency and quantity of the supplements used by the individual were recorded.

#### DETERMINATION OF ASCORBIC ACID LEVEL IN PLASMA

The validity of this questionnaire was evaluated by comparing the data obtained from the questionnaire with the level of ascorbic acid in plasma. In order to determine levels of ascorbic acid in plasma of individuals; following 12 hours of fasting, venous blood was collected from antecubital fossa using 10 ml vacuum tubes containing lithium heparin, was centrifuged for 10 minutes at 3000 rpm at 4°C, was sent to the laboratory for analysis. Determination of ascorbic acid in plasma was performed via high-performance liquid chromatography (HPLC).

#### STATISTICAL ANALYSIS

Statistical analyses were performed using Statistical Package for the Social Sciences (SPSS) 20.0 (SPSS Inc., Chicago, IL, USA) Software and Medcalc Software 12.4 (Medcalc Software Corp., Brunswick, ME, USA). Shapiro-Wilk test was used to evaluate normality of data distribution.<sup>19</sup> The median, 25<sup>th</sup> percentile, 75<sup>th</sup> percentile values of PAAL and vitamin C intake were shown. The relationship between PAAL and vitamin C intake was assessed by Spearman correlation test.<sup>20</sup> The classification into the same, adjacent and opposite quartiles between PAAL and intakes of vitamin C calculated by FFQ was evaluated by weighted kappa statistics. Also the Bland-Altman plots were used to compare two methods of assessments. The p value <0.05 was considered statistically significant.

## RESULTS

A total of 75 people (31 male and 44 female) participated in this study, and the mean age was 35.69±10.35 years (Table 1). The median PAAL was 7.94 mg/L, while the median intake of vitamin C was 136.84 mg/day (Table 2). There was a positive correlation between PAAL and intake of vitamin C calculated by FFQ (r=0.41; p<0.001) (Table 3).

The values of PAAL and intake of vitamin C calculated by FFQ were divided into quartiles; weighted kappa value were calculated for evaluating compatibility between two methods. The quartiles were shown in Table 4. The rate of the classifying into the same or adjacent quartile was 81.4%. The Bland-Altman plot was used to evaluate the compatibility between PAAL and intake of vitamin C calculated by FFQ (Figure 1).

**TABLE 1:** Demographic characteristics of the participants (n=75).

Characteristics	
Age (years), mean ± SD	35.69 ± 10.35
Gender, n (%)	
Male	31 (41%)
Female	44 (59%)
Education status, n (%)	
Below high school	24 (32%)
High school or above high school	51 (68%)
Marital Status, n (%)	
Married	36 (48%)
Single	39 (52%)
Average monthly income (TL), n (%)	
<2000	19 (25%)
2000-4000	42 (56%)
>4000	14 (19%)

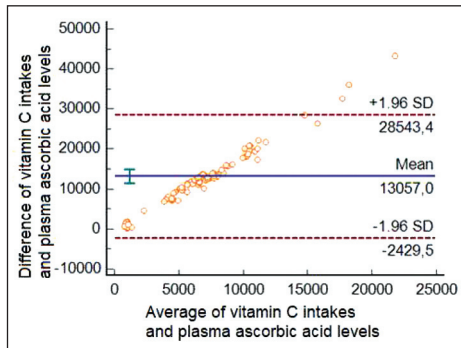
**TABLE 2:** The plasma ascorbic acid levels and intakes of vitamin C of individuals.

	Median	25 <sup>th</sup> percentile	75 <sup>th</sup> percentile
Plasma ascorbic acid level (mg/L)	7.94	5.69	10.64
Intake of vitamin C (mg/day)	136.84	114.00	187.80

**TABLE 3:** Relationship between plasma ascorbic acid level and intake of vitamin C.

	r	p <sup>†</sup>
Plasma ascorbic acid levels-	0.41	< 0.001
Intakes of vitamin C		

<sup>†</sup>Spearman correlation test.



**FIGURE 1:** Bland-Altman plots comparing the difference and means of vitamin C intakes and plasma ascorbic acid levels.

## DISCUSSION

In this study, an FFQ was developed to determine the level of dietary vitamin C intake and the validity of this FFQ was evaluated. Vitamin C intake was determined by FFQ and PAAL was determined by HPLC. The results were evaluated with Spearman correlation test, Bland Altman plot and kappa statistics. There are many factors that affect the relationship between the intake of vitamin C and the plasma level of vitamin C, such as food preparation/processing/storing/cooking, urinary excretion, sodium-dependent vitamin C transporter (SCVT)-1 and SCVT-2 genotypes, heavy metal poisoning, smoking or alcohol using.<sup>21,22</sup>

In the validation studies of FFQs, the relationship between FFQ and dietary record is generally examined. This value varies between studies. In the validation study of an FFQ developed by Gunes et

al., the correlation between dietary record and FFQ was investigated and the Pearson correlation coefficient was found to be 0.11 but in another study, this value was found as 0.98.<sup>23-25</sup> A FFQ was developed by Santosh and David to assess intake of vitamin C in Indian population. The validity of this questionnaire was evaluated by comparing with the results of 24 hour dietary record (p=0.03; r=0.99).<sup>26</sup>

Biochemical markers also can be used for validation of FFQs. Porrini and colleagues conducted the biochemical validation study of the FFQ they developed. As a result, they did not find a significant correlation between vitamin C intake and plasma ascorbic acid level (r=0.069; p=0676). Also, they divided the values of PAAL and vitamin C intake into quartiles, and the rate of the classifying into the same quartile was 36.8%. In our study, a significant correlation was found between two methods (r=0.41; p<0.001), and the rate of the classifying into the same quartile is higher (38.7%).<sup>27</sup>

Denghan et al. examined 20 different correlation values in their meta-analysis study; and found a positive correlation between the intake of vitamin C and PAAL (r value: 0.35 for all genders; 0.39 for females; 0.46 for males). It was expressed that this relationship was not strong, because of the effects of the bioavailability of vitamin C, the absorption conditions, the storage and processing conditions.<sup>12</sup> In this study, vitamin C content of diet was determined by the FFQ and validity of this FFQ was evaluated by comparison with the PAAL of the results and a positive correlation was found between two methods. Vitamin C, which is a water-soluble vitamin, is oxidized due to processes such as cutting, peeling, cooking and freezing. In addition, the serum / plasma ascorbic acid level decreases in cases such as severe trauma, stress, smoking, using of corticosteroid or oral contraceptives.<sup>8</sup> These factors affecting vitamin C content of

**TABLE 4:** Average values of plasma ascorbic acid levels and intakes of vitamin C calculated by FFQ were divided into quartiles, and weighted kappa values.

	The same quartile n(%)	Adjacent quartile n(%)	Opposite quartile n (%)	Kappa	p
Plasma ascorbic acid levels & intakes of vitamin C	29 (38.7%)	32 (42.7%)	14 (18.6%)	0.182	0.006



diet and the PAAL may have negatively affected the correlation examined in this study.

One of the limitations of this study was the low number of participants. Since this study was a biochemical validation, the number of participants could not be more. Another limitation was that all factors leading to loss of vitamin C could not be evaluated. The strengths of the study were that the cooking factor has been evaluated and this FFQ was the first questionnaire developed in our country to determine vitamin C intake.

## CONCLUSION

In conclusion, this is the first FFQ developed to determine vitamin C intake in Turkey, and has validity

coefficients similar to other FFQs. This FFQ can be used to determine intake of vitamin C.

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## Conflict of Interest

*No conflicts of interest between the authors and / or family members of the scientific and medical committee members or members of the potential conflicts of interest, counseling, expertise, working conditions, share holding and similar situations in any firm.*

## Authorship Contributions

*All authors contributed equally while this study preparing.*

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