Validation of protein and potassium intakes assessed from 24 h recalls against levels estimated from 24 h urine samples in children and adolescents of Turkish descent living in Germany: results from the EVET! Study

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Abstract

Objective: Nutrition-related health problems such as obesity are frequent among children and adolescents of Turkish descent living in Germany, yet data on their dietary habits are scarce. One reason might be the lack of validated assessment tools for this target group. We therefore aimed to validate protein and K intakes from one 24 h recall against levels estimated from one 24 h urine sample in children and adolescents of Turkish descent living in Germany.

Design: Cross-sectional analyses comprised estimation of mean differences, Pearson correlation coefficients, cross-classifications and Bland–Altman plots to assess the agreement between the nutritional intake estimated from a single 24 h recall and a single 24 h urine sample collected on the previous day.

Setting: Dortmund, Germany.

Subjects: Data from forty-three study participants (aged 5–18 years; 26% overweight) with a traditional Turkish background were included.

Results: The 24 h recall significantly overestimated mean protein and K intake by 10.7 g/d (95% CI of mean difference: 0.6, 20.7 g/d) and 344 mg/d (95% CI 8, 680 mg/d), respectively. Correlations between intake estimates were $r = 0.25$ ($P = 0.1$) and $r = 0.31$ ($P = 0.05$). Both methods classified 70% and 69% of the participants into the same/adjacent quartile of protein and K intake and misclassified 7% and 7%, respectively, into the opposite quartile. Bland–Altman plots indicated a wide scattering of differences in both protein and K intake.

Conclusions: Among children and adolescents of traditional Turkish descent living in Germany, one 24 h recall may only be valid for categorizing subjects into high, medium or low consumers.

Keywords

Validation
Protein intake
Potassium intake
24 h recall
Biomarkers
Children and adolescents of Turkish descent

People with a migrant background differ from people in their host country not only with respect to lifestyle factors but also in related health characteristics and disease frequency. In Germany, for example, a higher prevalence of overweight and obesity is observed among children and adolescents from families with Turkish origin1–3. The prevalence of diabetes mellitus type 2 amounts up to 14–29% in Turkish migrants living in Germany, whereas only 8–2% of people of German descent suffer from this disease4. According to the Federal Statistical Office, 6.8 million migrants live in Germany. Most persons with a migration background have belonged to German society for several decades; however, little is known about their lifestyle habits, especially with respect to health and nutrition.

To date, valid data describing food patterns of children from a Turkish background are scarce5. According to data from previous studies, Turkish children tend to consume relatively high amounts of raw vegetables, but also ingest less healthy foods like deep fried food and sweets more frequently than German children6–8. However, dietary assessment instruments, such as the FFQ used in these studies, permit only crude insights into dietary patterns of children and adolescents of Turkish descent.

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The FFQ method is highly culturally sensitive and tailored to its target population. Therefore, the results of such analyses are to be interpreted with caution. A dietary assessment instrument that is less sensitive to cultural differences is the 24 h recall. However, it has not been investigated how far 24 h recalls applied to this target group provide valid dietary data.

We therefore aimed to test the validity of a 24 h recall in a target group of children and adolescents of Turkish family background by comparing the reported intake data with urinary biomarkers: dietary intakes of protein and K as derived from a single 24 h recall were compared with the urinary excretion levels of protein and K. To that end we used data from the EVET! Study (Development, testing and validation of dietary assessment tools for children and adolescents of Turkish descent), which was conducted with the aim to develop, test and validate nutritional assessment tools for children and adolescents of Turkish descent in Germany.

Methods

The EVET! Study was conducted in the area of Dortmund, Germany, between March 2006 and September 2007. The German Research Institute of Child Nutrition conceived this study, funded by the German Federal Ministry of Food, Agriculture and Consumer Protection. All study procedures were approved by the Ethics Committee of the University of Bonn, Germany. Study participation was performed with parental consent or with the adolescent’s consent if the participant was 18 years old.

Study population

Study participants had to be of Turkish descent, aged 5–18 years and residents in the area of Dortmund, North Rhine-Westphalia, Germany. Generally, participants should have both parents from Turkish background; however, in one case the mother was of German ancestry and the father was of Turkish descent. Since successful recruitment turned out to be very difficult, various approaches were applied to recruit study participants for the EVET! Study such as direct invitation and indirect motivation via peer leaders. The complex methods of recruitment comprised the distribution of bilingual information on the study in locations such as schools, medical practices and one hospital, Turkish cultural associations, mosques and Turkish festivals. Finally, seventy-eight study participants, thirty-six (46%) male and forty-two (54%) female, could be included into the entire study. The present analysis considers data only of those children and adolescents who provided both a 24 h recall and a 24 h urine sample. Data from thirty-two participants had to be excluded due to incomplete or incorrect urine collections. In addition, three participants had provided 24 h recall data only with no concurrent 24 h urine sample. Hence, the final study population for the present analysis consisted of forty-three children and adolescents, fifteen (35%) male and twenty-eight (65%) female, aged 5–18 years.

Nutritional assessment

Dietary intake was assessed by a single 24 h recall based on EPIC-SOFT, a standardized computer program developed and validated by the International Agency for Research on Cancer in line with the EPIC (European Prospective Investigation into Cancer and Nutrition) study. Parents of children aged 5 to 10 years were interviewed in German or, if necessary, in Turkish, in the presence of their children, who provided additional information. Adolescents (11–18 years) were interviewed personally with their parents in the background in case of possible questions. Food and beverage consumption of the previous day was assessed, i.e. the day of the urine collection. The study participants quantified their food consumption by means of a picture book comprising German and Turkish food items, which was originally developed for EPIC-SOFT. Based on this book the EVET! Team added photos and descriptions of Turkish food portions. For the nutrient analysis of typical Turkish food items that were not available in the EPIC-SOFT nutrient database, the Turkish food database BeBiS (Beslenme Bilgi Sistemi, University of Istanbul) was used. Interviews were conducted in a standardized manner by trained and quality monitored bilingual personnel.

Urine collection and analysis

The 24 h urine collection, urine storage and analysis were performed according to the standard operating procedures of the DONALD (Dortmund Nutritional and Anthropometric Longitudinally Designed) Study. Parents and children were instructed personally and in written form in German and in Turkish how to collect complete 24 h urine samples.

The first micturition in the morning on the day of recording was discarded. Parents and children registered the time of this first micturition thereby defining the start of the 24 h collection, which ended with the first micturition of the following morning. All micturitions were stored immediately in Extran-cleaned, preservative-free (Extran MA 03; Merck, Darmstadt, Germany) 1-litre plastic containers with boracic acid at $\leq-12^\circ C$ before being transported to the research institute. A nutritionist (C.D.) picked up the urine samples, asked parents and children about the completeness of the urine samples and possible influences on urine collection, and recorded this information on the protocol sheet. At the institute, the containers were stored at $\leq-20^\circ C$ before being analysed. All urine samples underwent routine check using a commercial test strip after thawing and stirring. Volume, pH, osmolarity and creatinine were determined. Completeness of urine was ascertained via values of creatinine based on sex- and age-specific, body-weight-related reference values of creatinine for the age groups of 3, 4–5, 6–8, 9–13 and 14–18 years. Hence, collection-related
errors were identified. According to Remer et al.\(^{(16)}\) the lowest threshold for the excretion of creatinine in 24 h urine amounts to 0·1 mmol/kg per d, equivalent to the 5th percentile. Thus data on daily creatinine excretion rates that fell below 0·1 mmol/kg per d were regarded as incomplete and therefore not considered in our analysis.

Urinary N excretion was routinely measured by the method of Kjeldahl (Tecator 1002; Perstorp Analytical, Bristol, UK) and concentration was obtained by relating N levels to the volume of the 24 h collections. The assumption was made that excreted N accounts for 80 % of the ingested protein to account for extrarenal N losses\(^{(17,18)}\). Based on the molecular weight, excreted N (mmol/l) was converted to g protein/d, then multiplied by the factor 6·25, based on the assumption that all proteins contain 16 % N\(^{(19,20)}\), and then divided by 0·8\(^{(17,18)}\) to account for extrarenal losses. In a further step, an age- and sex-specific reference value of protein requirement for growth was added to allow for protein retention during growth in these paediatric age groups (see Table 1: protein intake estimated from 24 h urine allowing for retention)\(^{(21)}\).

For K intake, mmol/l was converted to g/d using molecular weight and then divided by 0·8 to account for an intestinal net absorption of 80 % for K intake\(^{(22,25)}\).

**Anthropometry and parental data**

Body weight was assessed to the nearest 100 g with an electronic scale, height to the nearest 0·1 cm and skinfold thickness to the nearest 0·1 mm. For each child, age- and sex-independent standard deviation scores of weight, height, BMI were calculated using the cut-off values proposed by the International Obesity Taskforce (IOTF)\(^{(24)}\). The proportion of overweight children was assessed according to the definitions of the IOTF, which correspond to a cut-off of 25 kg/m\(^2\) in adults\(^{(24)}\). On a child’s entry to school, parents were asked to provide information about family characteristics, their migration as well as educational and employment status.

**Statistical analysis**

Descriptive statistics of the study sample are presented as means and standard deviations or frequency and percentage. Mean differences and standard deviations were calculated\(^{(25)}\). By means of cross-classification, the percentage of persons who were either classified into the same or an adjacent quartile of protein or K intake by both methods or misclassified into the opposite quartile were computed\(^{(25)}\). Bland–Altman plots were used to illustrate the difference between both methods against the mean protein or K intake assessed by both methods\(^{(25–28)}\). The horizontal line indicates the mean of the differences. The upper and lower lines represent the upper and lower 95 % limits of agreement, which should comprise 95 % of the values in the range of the twofold standard deviation (1·96 × sd) of the mean differences, d (d ± 1·96 × sd). Ideally, the mean difference between the methods would be zero with no discernible bias, i.e. the mean differences would cluster on the horizontal line of equality (y = 0). Any deviation of the mean difference line from the line of equality indicates a bias. Moreover, any systematic variation of the differences in protein and K intake across the range of protein and K intakes suggests the presence of an additional systematic bias, which would provide further evidence of a limited agreement between the methods\(^{(26–28)}\). All statistical analyses were carried out using the SAS statistical software package version 8·2 (SAS Institute Inc., Cary, NC, USA) and P < 0·05 was considered statistically significant.

**Results**

The acceptability of collecting 24 h urine turned out to be low, thereby notably reducing the sample size available for the present analysis. Of the seventy-five urine samples collected, only forty-three conformed to the strict quality criteria established at the research institute. General characteristics of the available study sample are presented in Table 1. A relatively high prevalence of overweight (26 %) was observed. A higher educational level was seen among 26 % of the fathers and 14 % of the mothers. The prevalence of parental smoking was high (26 % and 61 % in mothers and fathers, respectively). Protein and K intake estimates were higher according to 24 h recalls than by 24 h urine collections.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>n or Mean</th>
<th>% or sd</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>43</td>
<td></td>
</tr>
<tr>
<td>% Female</td>
<td></td>
<td>65·0</td>
</tr>
<tr>
<td>Age (years)*</td>
<td>10–8</td>
<td>3·7</td>
</tr>
<tr>
<td>Overweight</td>
<td>11</td>
<td>25·6</td>
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<tr>
<td>Native language</td>
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<tr>
<td>German</td>
<td>3</td>
<td>7·0</td>
</tr>
<tr>
<td>Smoking (yes)</td>
<td>11</td>
<td>25·6</td>
</tr>
<tr>
<td>Higher education</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>14·0</td>
</tr>
<tr>
<td>Native language</td>
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<td></td>
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<tr>
<td>German</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Smoking (yes)</td>
<td>26</td>
<td>60·5</td>
</tr>
<tr>
<td>Higher education</td>
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<td></td>
</tr>
<tr>
<td></td>
<td>11</td>
<td>25·6</td>
</tr>
<tr>
<td>24 h recall*</td>
<td></td>
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<tr>
<td>Protein intake (g/d)</td>
<td>70</td>
<td>31</td>
</tr>
<tr>
<td>K intake (mg/d)</td>
<td>2263</td>
<td>1012</td>
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<tr>
<td>24 h urine collection*</td>
<td>Protein intake (g/d) estimated from 24 h urine allowing for retention</td>
<td>59</td>
</tr>
<tr>
<td>K intake (mg/d)</td>
<td>1926</td>
<td>774</td>
</tr>
</tbody>
</table>

*Values are represented as mean and sd.
†According to Cole et al.\(^{(26)}\).
‡Specific or general qualification for university entrance.
\(\dagger\)According to Garlick\(^{(27)}\).
\(\ddagger\)According to Remer and Manz\(^{(22,23)}\).
Table 2 presents the results of the validity analyses. Mean differences between both protein and K intake according to 24 h recalls and 24 h urine collections indicated an overestimation by the recall method. Pearson correlation coefficients were low for both protein and K intake. Only cross-classifications showed an acceptable agreement between the two methods: 70% and 69% of the participants were classified into the same or an adjacent category for protein and K intake, respectively. Misclassification was ≤7% in the total study group for protein as well as for K intake. The Bland–Altman plots indicated systematic overestimation of protein and K intakes and a large scatter of the differences for both protein (Fig. 1) and K (Fig. 2) due to wide confidence limits. Nevertheless, the plots do not suggest the presence of an additional systematic bias, since the differences between the measurements did not increase or decrease with increasing intake levels.

### Discussion

In this sample of children and adolescents with Turkish family background and low educational status living in Germany, a single 24 h recall proved to be of only limited

<table>
<thead>
<tr>
<th>Method of testing agreement</th>
<th>Protein intake (g/d)</th>
<th>K intake (mg/d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Differences</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absolute mean difference</td>
<td>+10.7</td>
<td>+344</td>
</tr>
<tr>
<td>95% CI</td>
<td>0.6, 20.7</td>
<td>7.6, 679.5</td>
</tr>
<tr>
<td>Pearson correlation coefficient, $r$</td>
<td>0.3</td>
<td>0.3</td>
</tr>
<tr>
<td>$P$ value</td>
<td>0.1</td>
<td>0.05</td>
</tr>
<tr>
<td>Cross-classification into quartiles</td>
<td>Same/adjacent</td>
<td></td>
</tr>
<tr>
<td>$n$</td>
<td>30</td>
<td>29</td>
</tr>
<tr>
<td>%</td>
<td>70.0</td>
<td>69.0</td>
</tr>
<tr>
<td>Opposite</td>
<td></td>
<td></td>
</tr>
<tr>
<td>$n$</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>%</td>
<td>7.0</td>
<td>7.0</td>
</tr>
</tbody>
</table>

Fig. 1 Bland–Altman plot for data of the total study group: children and adolescents (n 43) aged 5–18 years of Turkish descent living in Dortmund, Germany, 2006–2007 (EVET! Study). Difference between protein intake (g/d) calculated from a 24 h recall (test method) and the protein intake level (g/d) estimated from a 24 h urine sample (reference method) for each person (y-axis) plotted against the mean protein intake averaged from the two methods (x-axis). The mean of the differences is shown (---); also the upper and lower 95% limits of agreement (-----), presented as twofold standard deviations (± 1.96 × SD)
validity. When compared with urinary excretion data, the estimated intakes of both protein and K were considerably overestimated. Differences between the methods displayed a wide scatter and correlations were low. However, the ranking of subjects according to their intake levels appeared to be of acceptable validity.

To our knowledge, the present study is the first to validate protein and K intakes reported from 24 h recalls against protein and K intakes estimated from 24 h urines in a sample of healthy children and adolescents of Turkish descent living in Germany. Therefore, comparison with other study results is possible only to a limited extent. Similar studies in groups of children suffering from diabetes mellitus or renal insufficiency were conducted in non-migrant children only(29,30). One study reported an overestimation of only 2.29 g protein/d among 3–16-year old participants with diabetes mellitus, and observed higher correlation coefficients. However, similar to our study the authors concluded that the protein estimates from 24 h recall were satisfactory on the group level only(29,30). One study reported an overestimation of only 2.29 g protein/d among 3–16-year old participants with diabetes mellitus, and observed higher correlation coefficients. However, similar to our study the authors concluded that the protein estimates from 24 h recall were satisfactory on the group level only(29,30).

In our own previous study with data from the DONALD Study we observed a small but significant underestimation in protein intake of 6.31 g/d as derived from a 1 d weighed food record in comparison to excretion levels. However, the overall validity of the dietary assessment was considered acceptable(31). Unfortunately, data from 24 h recalls were not available to compare the validity between these two study populations.

The low number of comparable studies is probably due to the high level of compliance required from the children, adolescents and their parents for completion of both a dietary recall and a 24 h urine sample.

In contrast, validation studies in adults on 24 h recalls are more frequent(38,35,34). Pearson correlation coefficients of most of these studies were higher than those in our study, ranging from 0.46(33) to 0.94(34). Overestimation of protein intake by 1.31 g/d was observed in only one study(35), whereas other studies reported underestimations(31,33,34).

In the majority of cases, validation studies in children, adolescents and adults investigated other nutrients and/or validated nutrient intakes against nutritional assessment.
tools different from ours\textsuperscript{(32–37)}. Most of them found an acceptable validity of their tested instruments. However, none of them worked with a migrant population with comparatively low educational status, as was the case in the present study.

The reasons for the low validity of the applied dietary assessment methods observed in our study remain to be discussed. Clearly, this study population differed from most others in terms of their socio-economic and educational structure. Dietary assessment instruments in general are prone to biases due to educational attainment since they usually require a considerable ability to reflect on food intake, to oversee dietary habits over time, and they rely on certain knowledge on food and food preparation. Due to the usual recruitment of people with higher socio-economic status and with increased interest in diet and health, these other studies might have attained better validation results. Other factors influencing validity are sex, age and BMI\textsuperscript{(38,39)}. Furthermore, factors like socio-economic status, health-related activities or psychological factors are assumed to be determinants for misreporting, as suggested by a systematic literature review\textsuperscript{(40)}. In the present study, another source of under-reporting might have resulted from the specific interview situation: adolescents who were interviewed may have concealed information about food eaten away from the parents. In addition, parents may not have been able to report the complete food intake of their children due to food eaten away from home. As these facets are assumptions and as our study sample was very small, we could not evaluate how far these and possible other factors influenced our results.

It could be argued that the use of inappropriate assumptions for protein retention may have contributed to a lower validity for assessment of protein intake. However, the same assumptions did not result in a low validity in a previous analysis of data from the DONALD Study. Furthermore, results obtained for K in the present study were derived without such assumption, suggesting that in the present study the assessment itself is problematic rather than the calculations applied to the obtained data\textsuperscript{(19,32)}.

It is possible that repeated administration of 24 h recalls would result in more valid estimates of nutritional intake among children and adolescents of Turkish descent. However, the present study suggests that it may not be feasible to determine the validity of this alternative approach. Validation of repeated 24 h recalls would require repeated collections of 24 h urines, and 24 h urine collections were not accepted and/or well performed.

The major disadvantage of our study was the moderate coverage and response rate, which did not allow for a representative sample and stratified analyses. The major strength of our study was the recruitment of participants not often included in scientific investigations. The translation of all study documents into Turkish as well as the ability of our interviewers to perform 24 h recalls in Turkish language allowed us to include migrants of lower educational background compared with other investigations\textsuperscript{(41)}. Furthermore, our urine collection protocol was conducted analogously to that of the DONALD Study, known for its accuracy in urine sampling.

In summary, the results of our study suggest a low overall validity of protein and K intakes estimated from one single 24 h recall as evidenced from comparison with urinary excretion levels obtained from one 24 h urine sample. Only when based on group ranking level, a satisfactory validity could be observed. Therefore, in this population, a single 24 h recall should only be performed in order to rank participants into high, medium and low consumers.

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