

Validation of a Web-based, self-administered, non-consecutive-day dietary record tool against urinary biomarkers

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Abstract

New technologies are promising for the use of short-term instruments for dietary data collection; however, innovative tools should be validated against objective biomarkers. The aim of the present study was to investigate the validity of a Web-based, self-administered dietary record (DR) tool using protein, K and Na intakes against 24 h urinary biomarkers (24 h U). Of the total participants, 199 adult volunteers (104 men and 95 women, mean age 50.5 (23–83 years)) of the NutriNet-Santé Study were included in the protocol. They completed three non-consecutive-day DR and two 24 h U on the first and third DR days. Relative differences between reported (DR) and measured (24 h U) intakes were calculated from the log ratio (DR/24 h U) for protein, K and Na intakes: –14.4, +2.6 and –2.1% for men; and –13.9, –3.7 and –8.3% for women, respectively. The correlations between reported and true intakes were 0.61, 0.78 and 0.47 for men and 0.64, 0.42 and 0.37 for women for protein, K and Na, respectively. Attenuation factors, that represent attenuation of the true diet–disease relationship due to measurement error (a value closer to 1 indicating lower attenuation), ranged from 0.23 (Na, women) to 0.60 (K, men). We showed that the Web-based DR tool used in the NutriNet-Santé cohort study performs well in estimating protein and K intakes and fairly well in estimating Na intake. Furthermore, three non-consecutive-day DR appear to be valid for estimating usual intakes of protein and K, although caution is advised regarding the generalisability of these findings to other nutrients and general population.

Key words: Validation studies: Dietary records: Internet: Urinary biomarkers

Collection of high-quality dietary data in large populations is a challenging priority in nutritional epidemiology, in both aetiological research and surveillance studies. Bias due to measurement error of dietary factors is now widely acknowledged because no instrument to assess dietary intake is perfectly accurate^(1,2). Beyond unreliable descriptions of usual intakes, estimates of relationships between diet and disease may be attenuated or biased towards the null, and measurement error causes a loss of power to detect significant associations⁽³⁾.

The main dietary tools used in nutritional epidemiology are either contemporaneous dietary records (DR) or retrospective instruments such as multiple 24 h recalls or FFQ. Until recently,

repeated 24 h recalls or records on non-consecutive days were not used as main instruments for assessing diet in many cohort studies because of the substantial costs of repeated assessment to ensure reliable usual intake estimation. Instead, dietary exposure was mostly assessed through FFQ⁽⁴⁾, despite evidence that repeated 24 h recalls, taking into account the day-to-day variation, outperform FFQ in the accurate assessment of individual usual intake^(5–7).

The development of new technologies has led to an increasing number of innovative assessment tools, including online options, which are promising for applying in large-scale epidemiological studies^(8,9). In this context, Web-based

Abbreviations: 24 h U, 24 h urinary biomarkers; AMPM, Automated Multiple-pass Method; DR, dietary record; EFCOVAL, European Food Consumption Validation; OPEN, Observing Protein and Energy Nutrition; PABA, *para*-amino benzoic acid.

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self-administered tools for DR or 24 h dietary recalls could allow for accessing accurate dietary data on large samples with substantial resource savings. However, it is first necessary to validate such tools against objective markers of dietary intake.

'Recovery biomarkers' such as urinary N, K and Na are likely to closely reflect true dietary intake of these nutrients, and errors in measuring intake and urinary biomarkers are likely to be independent of each other⁽¹⁰⁾. This contrasts 'concentration biomarkers', such as plasma vitamins or fatty acids, which are subject to metabolic regulation and do not always correlate closely with intakes of their corresponding nutrients⁽¹¹⁾. Recovery biomarkers have been used in various dietary instrument validation studies (FFQ and 24 h recalls), including the OPEN (Observing Protein and Energy Nutrition) Study^(4,12), the EFCOVAL (European Food Consumption Validation) Study⁽¹³⁾, the Women's Health Initiative Nutritional Biomarker Study⁽¹⁴⁾ and the AMPM (Automated Multiple-pass Method)⁽¹⁵⁾, where the difference between reported and measured intakes could be estimated, as well as correlations between intakes and biomarker values.

NutriNet-Santé is the first Web-based, prospective cohort study that aims to investigate the relationship between nutrition and health⁽¹⁶⁾. Diet is assessed by three non-consecutive days of records at baseline and, again, at each year of follow-up. The dietary recording is self-administered through a specific Web-based tool, which has shown high agreement with an interview with a dietician as shown by median intra-class correlation and Pearson's correlation coefficients of 0.7–0.8⁽¹⁷⁾. However, this comparison study was not able to estimate the ability of the tool to assess true intake.

In the present study, we aimed to investigate the validity of a Web-based, self-administered DR tool of protein, K and Na intakes, as assessed by three non-consecutive DR days, against two non-consecutive measures of 24 h urinary biomarkers (24 h U) of these nutrients.

Materials and methods

Study population and ethics statement

The study participants were volunteers who participated in the NutriNet-Santé Study, an ongoing Web-based cohort study launched in France in May 2009, whose aims and methods have been described elsewhere⁽¹⁶⁾. Briefly, using a dedicated web site, adult volunteers (aged > 18 years) were followed for at least 10 years (recruitment still ongoing). Informed consent was obtained electronically from all participants. All procedures were approved by the International Research Board of the French Institute for Health and Medical Research (IRB Inserm no. 0000388FWA00005831) and the French National Information and Citizen Freedom Committee 'CNIL' (no. 908450 and 909216). At inception, participants completed a set of questionnaires assessing demographic, socio-economic and lifestyle factors, dietary intake measurements (three non-consecutive DR days), physical activity, anthropometry and health status. Dietary intake was evaluated again annually, and questionnaires on health status are sent on a regular basis.

A randomly selected sample of 1400 NutriNet-Santé Study participants living in Paris and greater area (for logistical reasons), stratified by sex, age (<45, >45 years) and educational level (primary and secondary up to some college, university graduate), were invited by e-mail to take part in the Dietary Validation Study. The objective was to recruit 200 participants. Since recovery biomarkers have been shown to be robust markers of dietary intake in individuals who are weight-stable and not experiencing illness⁽¹⁸⁾, exclusion criteria were as follows: self-reported metabolic disease (diabetes, heart failure, kidney failure or intestinal malabsorption, e.g. Crohn's disease); adherence to a weight-loss diet with observed weight loss > 1.5 kg/week over the past 4 weeks; currently pregnant or breast-feeding.

To ensure the validity of biomarkers derived from 24 h urine collections using *para*-amino benzoic acid (PABA), allergy to PABA was also an exclusion criterion. Participants were already enrolled in the NutriNet-Santé Study, and, thus, all had at least basic computer knowledge and no difficulty in understanding or reading French. The protocol was approved by the Consultative Committee Protecting Participants in Biomedical Research, Saint-Louis, Paris (no. 2011/22) and the 'CNIL' (DR-2012-467). Participants who completed the study received 100€ as compensation for the burdensome protocol.

Study design

Recruitment was carried out between October 2012 and April 2013. Interested subjects responded by e-mail and were subsequently contacted by telephone to check eligibility and to schedule their clinic visits and dates of DR and 24 h U. The study consisted of two visits at the clinical centre (Hôtel Dieu hospital, Paris), both in a fasting state (minimum of 6 h). At the first visit, clinical measurements were taken (blood pressure and heart pulse, height and weight). Participants were given instructions for the 24 h U collection and a physical activity questionnaire on occupational, transport and leisure time physical activity during the last 4 weeks to fill in at home (paper, self-administered) before the second visit. To complete the three DR days, a specific login and password was given to the participants. The second visit was scheduled approximately 3 weeks later. Between the two visits, three DR on non-consecutive days were self-administered through the specific Web-based tool. Two 24 h urine samples were collected per participant, covering the same 24 h periods as the first and the third DR days, with a time lag of approximately 2 weeks between the first and third DR. This scheme corresponds to the design participants follow in the NutriNet-Santé Study: three DR days randomly allocated for 2 weeks.

Dietary data collection

The Web-based tool was designed for self-administration and based on a secured user-friendly interface, designed by Medical Expert Systems©. Participants report all foods and beverages (type and quantity) consumed during all eating

occasions during 24 h from midnight to midnight. Participants first enter a list of every food item consumed at all eating occasions that they can recall via one of the following two ways: a food browser (foods are grouped by category) or a search engine that accepts spelling errors. Participants then estimate portion sizes of foods with the help of photographs, derived from a previously validated picture booklet that represents more than 250 generic foods⁽¹⁹⁾, corresponding to more than 2000 specific food items, presented in three different portions sizes. Along with the two intermediate and two extreme quantities, there are seven choices of amounts. Participants could also directly enter the quantity of foods consumed in grams or a measure of volume, use purchased units or describe intake in standard household units (e.g. teaspoons and tablespoons). Finally, after all food items and quantities have been entered, a summation is provided and participants have to review and describe if additional salt was consumed, and, if so, in what quantity (household units or grams). For each participant, daily nutrient intakes were calculated using the *ad-hoc* NutriNet-Santé composition table⁽²⁰⁾. An intake below 2092 kJ/d (500 kcal/d) for women or 3347 kJ/d (800 kcal/d) for men was considered implausible and excluded⁽²¹⁾, and the final analyses included only participants with at least two valid DR. Two DR were collected on weekdays and one on a weekend day.

24 h Urine collections and recovery biomarkers

At the first clinic visit, participants received instructions, materials (containers, four PABA pills) and a questionnaire for each 24 h urine collection. They were instructed to discard the first urine of the day of collection, and then to collect all urine passed during the next 24 h, up to the first urine passed on the next morning, which was also collected. During the day of collection, the container was kept at room temperature with the instruction to keep it in a dark place. To verify the collection samples, participants were asked to take two 100 mg PABA tablets on the day of collection and were informed that this process was to check the completeness of the collection as it may aid the collection of accurate samples⁽²²⁾. On the questionnaire, participants had to provide the times when collection started and finished (the following day), the time at which PABA pills were taken, any missing void and medications taken on that day. Urine samples were processed straight after collection the following morning: they were weighed, carefully mixed and aliquoted into 1 ml samples and stored at -80°C . In May 2013, all samples were transported to appropriate laboratories.

Urinary N concentration was measured by pyrochemoluminescence on an Antek 9000 analyzer, which produces results very well correlated with the reference method (the Kjeldahl technique)⁽²³⁾, at Cochin Hospital, Université Paris Descartes. K and Na concentrations were measured by ion-selective electrodes (Siemens Dimension Vista) at the laboratory of Nutrition Hormonology in the CHU (Centre Hospitalier Universitaire) of Grenoble. Creatinine concentration, used as a marker to check for validity of urine collection, was measured by alkaline picrate kinetic (Siemens Dimension

Vista) also in Grenoble. The CV of these analyses (intra-assay precision) was $<3\%$.

Covariate assessment

Height of the participants was measured without shoes to the nearest 0.5 cm by a trained technician, using a wall-mounted stadiometer⁽²⁴⁾. Weight (to the nearest 0.1 kg) of the participants (wearing only underwear) was measured with a calibrated impedance body composition analyzer (BC-418MA; Tanita®). BMI was calculated as the weight (kg) divided by the squared height (m^2). Dietary supplement use, frequency and type were determined by a questionnaire.

Statistical analysis

Description of the characteristics of the study participants (means and standard deviations or n and %) was compared between men and women using Kruskal–Wallis (when normality was not met) or t test for continuous variables and χ^2 tests for categorical variables.

Assuming that approximately 81% of N is excreted via urine in 24 h, and that proteins contain 16% of N⁽²⁵⁾, that 77% of K⁽²⁶⁾ and 86% of Na⁽¹⁵⁾ are excreted in 24 h, we could calculate the biomarker-based intakes:

$$\text{Protein (g/d)} = \text{urinary N (mol/l)} \times \text{volume 24 h U (litres)} \times 14 \text{ (g/mol)} \times 6.25/0.81,$$

$$\text{K (mg/d)} = \text{urinary K (mol/l)} \times \text{volume 24 h U (litres)} \times 39 \text{ (g/mol)} \times 1000/0.77,$$

$$\text{Na (mg/d)} = \text{urinary Na (mol/l)} \times \text{volume 24 h U (litres)} \times 23 \text{ (g/mol)} \times 1000/0.86.$$

24 h Urine collections were determined as valid using the following criteria: collection time between 22 and 26 h; urine volume $\geq 500 \text{ ml}^{(15)}$; reported missing urine (estimated volume missed void $>5\%$ total volume) and creatinine >10 or $>15 \text{ mg/kg}$ for women and men, respectively⁽²⁷⁾. If one or more of the listed criteria was not met, then the 24 h U collection was considered invalid. The following sensitivity analyses were conducted: (1) exclusion of urine samples with >1 reported missing void because people admitting one missing void might be actually more diligent or have missed only a small volume compared with those reporting more than one missing void⁽²²⁾; (2) exclusion of participants with one invalid urine measure.

All intake and excretion values were log-transformed to improve normality. Intra-cluster correlation coefficients between U1 and U2 (using the mean of two measurements), and between the three DR (using the mean of three measurements), were calculated with the SAS macro %ICC9⁽²⁸⁾. Mean protein, K and Na intakes based on up to 3 d of DR (R_{ij} for an individual i on a day j), and excretion on up to 2 d of 24 h U (M_{ij}), were calculated on the log-transformed values and exponentiated to obtain geometric means and 95% CI. For an individual, the log-ratio $\log(\bar{R}_i/\bar{M}_i)$ was calculated, where \bar{R}_i is the individual mean of up to three DR and \bar{M}_i is

the mean of up to two 24 h U. After exponentiation of the sample mean log-ratio, with a ratio of 1 representing no difference between intake and excretion, we expressed the distance to the reference in percent, e.g. a ratio of 0.90 (90%) is equivalent to a relative difference of -10%. Misreporting refers to the presence of a significant difference.

A ratio below 70% indicated the presence of severe under-reporting, between 70 and 80% moderate under-reporting, between 80 and 120% correct reporting, and above 120% over-reporting bias⁽²⁹⁾.

We calculated the ratio across age categories (≤ 45 and > 45 years) and across BMI categories (< 25 , $25-29.9$, and ≥ 30 kg/m²) and compared them using ANOVA after assumptions were checked.

To assess the validity of the DR tool, we calculated Pearson's correlation coefficients and their confidence interval using the Fisher's Z transformation, both unadjusted and adjusted for age, BMI, physical activity and energy intake (by the residual method⁽²¹⁾).

To examine the structure of the measurement errors, a complex measurement error was assumed⁽¹⁰⁾. It is described in online Supplementary material. This allowed for the calculation of the correlations between reported and true intakes on the same given day (assesses if the instrument measures what it is supposed to be measured), and correlation coefficients between usual reported intake and true intake, as well as attenuation factors (λ)⁽¹⁰⁾. Attenuation factors represent the attenuation of the strength of the relationship between nutrient intake and a disease of interest; a value closer to 1 meaning that there is less attenuation (with 1 representing no attenuation at all). Although no exact cut-off exists to interpret correlation and attenuation coefficients, a value of at least 0.40 would avoid needing hugely inflated sample sizes to observe significant diet-disease relationship⁽³⁰⁾; hence, values ≥ 0.40 were deemed acceptable/fair, ≥ 0.60 as high and < 0.40 as low.

All analyses were performed using SAS version 9.3 (SAS Institute, Inc.), the significance level was two-sided and set at $P=0.05$.

Results

Subject characteristics

Of the 1400 individuals contacted by e-mail, 237 (16.9%) responded. Of these, seven (3%) were ineligible and thirty-one (13%) were not able to attend the planned clinic visits; hence, 199 participants were included in the study.

A total of 398 24 h U specimens were available. Both 24 h U measurements were invalid for four female participants and one male participant; hence, these five participants were excluded from the analysis. One man had one invalid 24 h U and two implausible DR, and was thus excluded. Finally, 193 subjects were included in the analysis. Twenty-five subjects had data for only one 24 h U because fourteen (7.3%) first 24 h U and eleven (5.7%) second 24 h U were considered invalid.

Participant characteristics are presented in Table 1. The sample was composed of 47.7% of females, who did not differ from males in terms of age (50.5 (SD 16.4) years) or

BMI (24.0 (SD 3.5) kg/m²). Obesity (BMI ≥ 30 kg/m²) was more common in women than in men (12 *v.* 3%); however, overweight (25 \leq BMI < 30 kg/m²) was more common in men (36 *v.* 18%). Women had a higher frequency of dietary supplement use (36 *v.* 24%). Men had higher energy intake (10 000 kJ in men *v.* 7172 kJ in women). Energy from protein was lower for men than women; however, energy from fat and carbohydrates were not appreciably different.

Intakes of protein, potassium and sodium and misreporting

Intakes of protein, K and Na based on three DR days and two 24 h U excretion are summarised in Table 2. Intra-cluster correlation coefficients between U1 and U2 were 0.60 for proteins, 0.45 for K and 0.36 for Na, and between three diet records 0.52, 0.54 and 0.47 for protein, K and Na, respectively.

Men and women under-reported their protein intake (-14.4 and -13.9%, respectively, NS between-sex difference, $P=0.88$). Men showed non-significant over-reporting for K and Na intakes, while women under-reported these two nutrients.

Misreporting was greater in women aged > 45 years than those aged ≤ 45 years for intakes of protein (-17 *v.* -8%, $P=0.047$) and Na (-15 *v.* +3%, $P=0.04$); however, no significant difference across age categories was observed for K, and no misreporting differences were observed for males. By BMI categories, misreporting of Na intake was greater for obese women than overweight or normal-weight men, although the difference did not reach statistical significance (-25% in obese, -2% in overweight and -7% in normal weight, $P=0.13$).

The frequency of misreporting is summarised in Table 3. The difference between men and women was non-significant; however, a trend was observed for K with more men over-reporting (24.5%) than women (20.9%), and for Na with more women severely under-reporting (29.7%) than men (16.7%).

Correlations and attenuation

Correlation coefficients between intake (DR) and excretion (24 h U) are summarised in Table 4. Higher correlations were observed for men than for women for all the three nutrients. Crude correlations ranged from 0.45 (Na) to 0.63 (K) for men, and from 0.27 (Na) to 0.54 (protein) for women. Adjusted correlations for age, BMI, level of education and energy intake were higher than the crude coefficients for women, but lower for men.

Sensitivity analyses taking into account only the first and third DR, which correspond to the days of 24 h U collection, showed overall similar results for relative differences and correlations; the only notable exception was a lower correlation between Na intake and excretion in men (r 0.17).

Taking into account the complex measurement error model, we calculated the correlations between reported intake by one DR and true intake on the same day (Table 5). These coefficients were higher than crude correlations for women, and similar to those for men.

Finally, correlations between intake of the average of three DR and true usual intake (Table 6) were high for protein in



Table 1. Characteristics of participants in the NutriNet-Santé Dietary Validation Study, France, 2013

(Number of participants and percentages; mean values and standard deviations)

	Men (n 104)		Women (n 95)		P*
	n	%	n	%	
Age (years)					0.9
Mean	50.3		50.7		
SD	16.1		16.8		
Median	51		54		
Q1–Q3	35–65		35–65		
BMI (kg/m ²)					0.6
Mean	24.1		23.9		
SD	2.9		4.2		
Weight (kg)					<0.0001
Mean	74.8		62.7		
SD	10.9		10.7		
Height (cm)					<0.0001
Mean	176.0		162.3		
SD	7.1		6.0		
Physical activity (MET-h/week)					0.6
Mean	85.1		81.6		
SD	48.7		50.1		
LTPA (MET-h/week)					0.0002
Mean	35.7		21.4		
SD	29.9		21.9		
Use of dietary supplement	25	24.0	34	35.8	0.07
BMI category (kg/m ²)					0.001
Underweight (<18.5)	1	1.0	7	7.4	
Normal (18.5–24.9)	63	60.6	60	63.2	
Overweight (25–29.9)	37	35.6	17	17.9	
Obese (≥30)	3	2.9	11	11.6	
Tobacco smoking					0.35
Smoker – regularly	9	8.7	10	10.5	
Smoker – occasionally	3	2.9	6	6.3	
Former smoker	39	37.5	26	27.4	
Never smoker	53	51.0	53	55.8	
Living with a partner	69	66.3	53	55.8	0.13
Occupation					0.04
Never employed	3	2.9	6	6.3	
Self-employed farmers	2	1.9	1	1.1	
Managerial/professional position	45	43.3	30	31.6	
Manual workers	1	1.0	0	0.0	
Blue collar	15	14.4	27	28.4	
Retired	38	36.5	31	32.6	
Education					0.10
Up to high school	21	20.2	18	18.9	
Some college	34	32.7	25	26.3	
University graduate	49	47.1	52	54.7	
Dietary intake†					
Energy (kJ)					<0.0001
Mean	9999.8		7172.2		
SD	2536.8		1735.9		
Carbohydrate density‡					0.31
Mean	42.2		41.2		
SD	6.7		6.9		
Protein density§					0.03
Mean	16.7		17.8		
SD	3.5		3.8		
Lipid density§					0.86
Mean	40.9		40.7		
SD	6.6		6.9		
Alcohol (g)					0.001
Mean	13.9		7.3		
SD	16.4		8.6		
Dietary fibre (g)					0.0001
Mean	24.5		20.0		
SD	9.7		6.0		

MET, metabolic equivalents of task; LTPA, leisure time physical activity.

* The difference between men and women was estimated using *t* test and χ^2 tests as appropriate.

† Mean intake was calculated from three non-consecutive dietary record days.

‡ Percentage of energy intake (excluding alcohol).

Table 2. Intake of protein, potassium and sodium from non-consecutive-day dietary records (DR) and 24 h urine excretions, NutriNet-Santé Dietary Validation Study, France 2013

(Geometric mean values and 95 % confidence intervals)

	Men (n 102)			Women (n 91)			P*
	n	Geometric mean	95 % CI	n	Geometric mean	95 % CI	
Protein (g/d)							
24 h U 1	96	104.8	61.4, 179.0	86	82.9	49.0, 140.3	<0.0001
24 h U 2	97	102.4	62.3, 168.2	82	76.6	41.1, 142.8	<0.0001
Mean 24 h U	102	101.7	62.3, 166.2	91	77.4	45.8, 130.5	<0.0001
24 h DR 1	102	90.3	84.5, 96.5	91	70.3	66.1, 74.7	<0.0001
24 h DR 2	102	88.9	82.7, 95.6	90	68.4	63.1, 74.0	<0.0001
24 h DR 3	101	86.9	81.2, 93.1	90	67.6	63.2, 72.4	<0.0001
Mean 24 h DR	102	88.6	83.9, 93.7	91	68.8	65.1, 72.8	<0.0001
Difference %†	102	-14.4	-18.2, -10.3	91	-13.9	-18.3, -9.3	0.88
K (mg/d)							
24 h U 1	96	3407	3210, 3616	86	3012	2814, 3224	0.008
24 h U 2	97	3353	3165, 3552	82	2672	2486, 2872	<0.0001
Mean 24 h U	102	3357	3189, 3535	91	2843	2685, 3010	<0.0001
24 h DR 1	102	3468	3266, 3683	91	2800	2624, 2988	<0.0001
24 h DR 2	102	3490	3279, 3714	90	2684	2530, 2847	<0.0001
24 h DR 3	101	3379	3191, 3577	90	2717	2545, 2900	<0.0001
Mean 24 h DR	102	3444	3279, 3618	91	2739	2607, 2879	<0.0001
Difference %†	102	2.6	-1.7, 7.1	91	-3.6	-8.9, 1.9	0.08
Na (mg/d)							
24 h U 1	96	3667	3355, 4007	86	3105	2855, 3377	0.009
24 h U 2	97	3576	3295, 3881	82	2836	2581, 3118	0.0003
Mean 24 h U	102	3578	3320, 3856	91	2996	2790, 3217	0.001
24 h DR 1	102	3600	3308, 3918	91	2812	2580, 3065	<0.0001
24 h DR 2	102	3503	3195, 3841	90	2703	2467, 2962	0.0001
24 h DR 3	101	3411	3139, 3706	90	2706	2485, 2948	0.0002
Mean 24 h DR	102	3503	3271, 3752	91	2747	2567, 2941	<0.0001
Difference %†	102	-2.1	-9.2, 5.6	91	-8.3	-15.7, -0.2	0.26

24 h U, 24-h urine collection.

*The difference between men and women was estimated using *t* test.

†Mean difference in % was calculated from the log ratio of mean reported intake (non-consecutive DR) over mean biomarker intake (24 h U (24 h urinary

biomarkers)) following the formula $100 \left[\exp \left(\frac{\sum_{i=1}^n \log \left(\frac{\bar{R}_i}{\bar{M}_i} \right)}{n} \right) - 1 \right]$, where \bar{R}_i is the geometric mean of DR for an individual *i* across the three

measurements; \bar{M}_i is the geometric mean of 24 h U for an individual *i* across the two measurements; and *n* the number of individuals in the sample. A mean log ratio of zero would represent no difference in reporting compared with the biomarker measure. The exponentiation allows to express it as a ratio whose reference value is 1, and we further expressed it as a per cent difference, e.g. a ratio of 0.90 is a per cent difference of -10%.

both men and women (>0.60), very high for K in men, while only fair in women, and fair (men) to poor (women) for Na. Attenuation factors ranged from 0.23 (Na, women) to 0.60 (K, men).

Discussion

The present validation study is the first to examine the structure of the measurement error with repeated Web-based, self-administered, non-consecutive-day DR, allowing for the estimation of the correlations with true intakes of protein, K and Na. Only a few studies^(4,13,29,31-35) have assessed the validity of repeated short-term instruments, such as 24 h recalls, against biomarkers, and none has validated Web-based self-administered non-consecutive DR.

Misreporting of protein, potassium and sodium intakes

We found that on average, men under-reported protein but slightly over-reported their K and Na intake, whereas women under-reported protein, K and Na intakes. Correlation coefficients indicated that three non-consecutive 24 h diet records self-administered via the Web-based tool perform

well for the estimation of protein and K intakes, and fairly well for estimating Na intake.

The EFCOVAL and the OPEN studies aimed to validate two 24 h recalls, administered by a dietitian, against urinary biomarkers. Results in the French EFCOVAL centre showed under-reporting of -12.1% for protein and -17.1% for K in men and -12.8 for protein and -13.0% for K in women, respectively⁽¹³⁾. For protein, the results are similar to our findings; however, for K, under-reporting was much more prominent in the EFCOVAL Study than in the present study. In the American OPEN Study, under-reporting of protein was also similar (-11 to -12%)⁽⁴⁾. Regarding Na, the United States Department of Agriculture AMPM validation study⁽¹⁵⁾, with two 24 h urine collections covering the same time period as two 24 h recalls, showed greater under-reporting (-7% for men and -10% for women) than in the present study. Protein, K and Na find their main source in very different food groups, and represent different aspects of diet quality, so it is not surprising that dietary misreporting differs across nutrients, as suggested elsewhere⁽³⁶⁾.

Crude correlation coefficients in EFCOVAL Study were 0.65 (protein) and 0.62 (K) in men and 0.46 (protein) and 0.61 (K) in women, respectively, which is slightly higher than those in

Table 3. Frequency of misreporting* in protein, potassium and sodium intakes, NutriNet-Santé Dietary Validation Study, France 2013

(Number of participants and percentages)

	Men (n 102)		Women (n 91)		P†
	n	%	n	%	
Protein					0.88
Over-reporter	7	6.9	9	9.9	
Correct reporter	58	56.9	51	56.0	
Moderate under-reporter	17	16.7	13	14.3	
Severe under-reporter	20	19.6	18	19.8	
K					0.22
Over-reporter	25	24.5	19	20.9	
Correct reporter	63	61.8	51	56.0	
Moderate under-reporter	11	10.8	12	13.2	
Severe under-reporter	3	2.9	9	9.9	
Na					0.19
Over-reporter	30	29.4	24	26.4	
Correct reporter	38	37.3	26	28.6	
Moderate under-reporter	17	16.7	14	15.4	
Severe under-reporter	17	16.7	27	29.7	

* Based on the log ratio of mean reported intake (non-consecutive DR) over mean biomarker intake (24h U (24h urinary biomarkers)). Ratio <70%: severe under-reporter; 70% < ratio < 80%: moderate under-reporter; 80% < ratio < 120%: normo-reporter; ratio > 120%: over-reporter.

† The difference between men and women was estimated using Fisher's exact test.

the present study. However, correlation coefficients for protein found in the present study are somewhat higher than usually reported in other validation studies including short-term instruments (24h recalls), such as the OPEN Study (r 0.41 for men and r 0.26 for women)⁽⁴⁾, the DEARR (Dietary Evaluation and Attenuation of Relative Risk) study (r 0.29)⁽³⁴⁾ or the UK arm of EPIC (European Prospective Investigation into Cancer and Nutrition) (r 0.10 for one 24h recall)⁽³¹⁾, and are more similar to the one observed with a 7-d diary (r 0.65)⁽³¹⁾.

Greater misreporting and lower correlation coefficients for all the three nutrients (protein, K and Na) were observed in women than in men in the present study, which is fairly consistent with most of the validation studies of short-term instruments for protein^(4,13), K⁽¹³⁾ or Na⁽¹⁵⁾. Although the present study does not allow exploring this aspect in depth, differences in social desirability is a potential explanation because of the societal pressure placed on women to be slim. Women, more than men, may under-report to prevent being seen as indulging in an undesirable behaviour, such as eating unhealthy food or overeating^(37,38).

We found no significant difference in misreporting of protein, K or Na according to BMI categories. However, for protein, the trend was towards more under-reporting of intake among the overweight or obese individuals than among the normal-weight individuals. Given the very low number of obese men (n 3) in the study, we carried out the analyses between normal-weight (BMI <25 kg/m²) and overweight/obese (BMI ≥25 kg/m²) men and showed the same non-significant trend (−18% in overweight *v.* −12% in normal weight, $P=0.16$). This follows the trend observed in the OPEN Study: lower correlation coefficients between reported protein intake (average of two 24h recalls) and biomarkers in obese than in non-obese men (r 0.217 *v.*

0.483, $P=0.05$)⁽¹²⁾. For K, BMI classification did not seem to influence misreporting. For Na, the AMPM validation study found that overweight and obese men and women under-reported more than their normal-weight counterparts. This finding is similar to the trend observed in the present study for women. Across age categories, in the AMPM, females under 50 years tended to under-report Na intake more than their elder counterparts (−15 *v.* −5%) whereas we found the opposite. This can be explained by a lower computer knowledge among the older participants⁽³⁹⁾, and these results are consistent with those of the comparison study of our tool with a 24h recall assessment by a dietitian, where the proportion of 'novice or inexperienced with computer' was higher among women than men⁽¹⁷⁾.

Besides, it is known that dietary misreporting (particularly energy under-reporting) is more frequent among the elderly^(40,41). The present study includes six participants aged ≥75 years (three men and three women). When we excluded them from the main analysis, the results remained unchanged. However, among these six participants, we observed greater under-reporting of K (−13.4% in men and −14.6% in women), protein for men (−19.3%) and Na for women (−36.8%), although the Kruskal–Wallis test showed no significant difference (all $P>0.05$), which is likely to be due to a lack of power. These results may imply that extra attention should be paid to the quality of dietary data when studying diet–disease associations among the elderly.

Correlations with true intake and structure of the measurement error

Correlations between reported intake and true intake were not estimated in the EFCOVAL or AMPM studies, but were estimated in the OPEN Study⁽⁷⁾. It was estimated that four 24h recalls could lead to a correlation coefficient of 0.508 (men) and 0.440 (women) with true intake of protein. The correlations between the average of three non-consecutive-day records and true intake observed in the present study

Table 4. Pearson's correlation coefficients (r) between reported intakes by three non-consecutive-day dietary records and excretions in two 24h U (24h urinary biomarkers) for protein, potassium and sodium intakes, NutriNet-Santé Dietary Validation Study, France 2013

(Pearson's correlation coefficients and 95% confidence intervals)

	Men (n 102)		Women (n 91)	
	r	95% CI	r	95% CI
Protein				
Unadjusted	0.61	0.47, 0.72	0.54	0.37, 0.67
Adjusted*	0.56	0.41, 0.68	0.55	0.39, 0.68
K				
Unadjusted	0.63	0.50, 0.74	0.45	0.27, 0.60
Adjusted*	0.62	0.48, 0.73	0.51	0.33, 0.65
Na				
Unadjusted	0.45	0.28, 0.59	0.27	0.06, 0.45
Adjusted*	0.31	0.12, 0.48	0.34	0.14, 0.52

* Pearson's correlation adjusted for energy intake by the residual method, age, BMI and level of education.

Table 5. Estimated correlation (*r*) between one dietary record (DR) and true intake on the same day for protein, potassium and sodium intakes, NutriNet-Santé Dietary Validation Study, France 2013

(Estimated correlation coefficients and 95% confidence intervals)

	Men (n 102)		Women (n 91)	
	<i>r</i> *	95% CI	<i>r</i> *	95% CI
Protein	0.60	0.46, 0.75	0.59	0.41, 0.76
K	0.68	0.53, 0.83	0.51	0.25, 0.77
Na	0.45	0.26, 0.63	0.39	0.11, 0.66

* Correlation coefficient between DR and true intake on the same given day as estimated by the model accounting for the reference biomarkers (24h U (24h urinary biomarkers)) as reference measurement. For more details on calculation, see online Supplementary material.

(0.61 in men and 0.64 in women) are higher and actually outperform the prediction by Schatzkin *et al.*⁽⁷⁾ with a theoretically infinite number of 24 h recalls (0.597 for men and 0.584 for women).

Attenuation factors found in the present study are similar to the estimates from four 24 h recalls in the OPEN Study for protein in men (0.37), and higher in women (0.43 in our study *v.* 0.32 in OPEN Study)⁽⁷⁾, a higher value indicating less bias in estimating diet–health relationships. For K, we found a higher attenuation factor for men, i.e. less bias, than in the OPEN Study (0.60 *v.* 0.32), but a slightly lower factor for women (0.29 *v.* 0.33)⁽⁴⁾. No comparison can be made for Na since, to our knowledge, no other study has estimated attenuation factors for this nutrient.

Finally, this is the first study to assess the correlations between Web-based, self-reported and true intake on a given day, which is a method for evaluating how well the instrument measures its target, without penalising the correlation for the fact that dietary intake may exhibit considerable daily variability. The correlation coefficients were high for protein in both sexes, high for K in men and fair in women, and fair for Na in both men and women. Coefficients were lower for women than men, indicating a lower intrinsic validity of the instrument for women than for men.

Methodological considerations

The main strength of the present study is the use of objective biomarkers, namely 24 h U protein, K and Na, collected on the same day of diet record, in a repeated fashion, which allowed for the estimation of the extent of misreporting, as well as same-day correlations and for usual intake with a complex measurement error model. Accuracy – i.e. completeness – of the 24 h urine collections was assessed comprehensively by different criteria: creatinine (five invalid), total volume (one invalid); self-report of missing voids (twenty-three invalid). Also, although PABA was not assayed, participants were asked to take the PABA pills during the collection which potentially has a ‘placebo effect’ to engage in more compliant behaviour⁽²²⁾. Results of both sensitivity analyses using different criteria for exclusion were identical for women; however, slightly lower correlation coefficients and

attenuation factors were observed for men. This seems to imply that our strategy of exclusion of invalid urine was an adequate balance between accuracy and statistical power.

Finally, as our strategy of excluding DR days with implausibly low energy intake may introduce bias, we repeated the analyses including the three implausible DR, which did not change the results.

The main limitation of the present study is the absence of use of a recovery biomarker for energy intake, namely double-labelled water, which requires a much more costly and burdensome protocol. Hence, although protein intake, given its energy content, can be used as a proxy of energy intake, we cannot extrapolate the results on protein intake to other macronutrients or total energy intake, as suggested by the OPEN Study results^(4,10). An important issue in validating dietary assessment tool is the current paucity of valid recovery biomarkers; however, emerging food metabolomics studies may be a promising way to assess nutritional intake through biomarkers⁽⁴²⁾.

Caution is advised when extrapolating from the results of the present validation study to the general population because it was carried out on a relatively small sample of subjects. These were volunteers and probably differed in terms of socio-economic, demographic and lifestyle characteristics from the general population. However, we carried out our sampling strategy in order to have a wide spectrum of age, education level and equal numbers of men and women so that validity could be assessed irrespective of these parameters.

We showed that the Web-based, repeated, non-consecutive-day DR tool used in the NutriNet-Santé cohort study performs well in estimating protein and K intakes and fairly well in estimating Na intake. Furthermore, three repeated DR appear to be valid to estimate usual intakes of protein and K, although caution is advised regarding the generalisability of these findings to other nutrients and to the general population.

Table 6. Estimated correlation (*r*) between the average of three non-consecutive-day dietary record (DR) and true usual intake and attenuation factor (Atten) for protein, potassium and sodium intakes, NutriNet-Santé Dietary Validation Study, France 2013

(Estimated correlation coefficients, attenuation factors and 95% confidence intervals)

	Men (n 102)		Women (n 91)	
	<i>r</i> *	95% CI	<i>r</i> *	95% CI
Protein	0.61	0.43, 0.78	0.64	0.43, 0.85
K	0.78	0.61, 0.94	0.42	0.13, 0.71
Na	0.47	0.23, 0.71	0.37	0.03, 0.70
	Atten†	95% CI	Atten†	95% CI
Protein	0.37	0.24, 0.50	0.43	0.26, 0.59
K	0.60	0.44, 0.76	0.29	0.06, 0.52
Na	0.37	0.17, 0.56	0.23	0.01, 0.45

* Correlation coefficient between the average of three non-consecutive-day DR and true usual intake as estimated by the model accounting for the reference biomarkers (average of three 24h U (24h urinary biomarkers)) as reference measurement.

† Interpretation of attenuation factor: a value closer to 1 indicates lower attenuation of the true relationship between intake and disease. For more detail on calculation, see online Supplementary material.

Supplementary material

To view supplementary material for this article, please visit <http://dx.doi.org/10.1017/S0007114515000057>

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Details of measurement error models are available as online supplementary material with the online posting of this paper.

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The authors' contributions are as follows: C. L., E. K.-G., K. C., V. D., M. V., P. G. and S. H. were responsible for developing the design and protocol of the study; C. L. conducted the research, carried out data checking and analyses, and was responsible for drafting the manuscript; K. C., V. D., M. V., G. M. C., P. G., S. H., E. K.-G., F. L. and P. F. were involved in interpreting the results and editing of the manuscript; F. L. and P. F. carried out the biomarker analyses. All authors read and approved the final manuscript.

The authors declare that there are no conflicts of interest.

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