International Journal of Epidemiology
Vol. 26, No. 1(Suppl. 1)

Printed in Great Britain

Validation of Dietary Assessment Methods in the UK Arm of EPIC Using Weighed Records, and 24-hour Urinary Nitrogen and Potassium and Serum Vitamin C and Carotenoids as Biomarkers

SHEILA A BINGHAM,* CAROLINE GILL, AILSA WELCH,** AEDIN CASSIDY,* SHIRLEY A RUNSWICK,* SUZY OAKES,† ROBERT LUBIN,** DAVID I THURNHAM,‡ TIMOTHY J A KEY,§ LYNN ROE,‖ KAY-TEE KHAW† AND NICHOLAS E DAY****

Bingham S A (M R C Dunn Clinical Nutrition Centre, Hill Road, Cambridge, UK), Gill C, Welch A, Cassidy A, Runswick S A, Oakes S, Lubin R, Thurnham D I, Key T J A, Roe L, Khaw K-T and Day N E. Validation of dietary assessment methods in the UK arm of EPIC using weighed records, and 24-hour urinary nitrogen and potassium and serum vitamin C and carotenoids as biomarkers. *International Journal of Epidemiology* 1997; **26 (Suppl. 1):** S137–S151.

Background. In the UK EPIC validation studies, the accuracy of several methods was assessed by comparison with 16-day weighed records and the biomarkers, 24-hour urine nitrogen (N) and potassium (K), plasma carotenoids and plasma vitamin C.

Methods. Comparisons between methods were made on 156 women, studied over 1 year at 3-monthly intervals at home. On each of four occasions, volunteers completed 4 days of weighed records and provided two 24-hour urine collections and a fasting blood sample.

Results. In comparison with the 16 days of weighed records, a food frequency questionnaire (FFQ) yielded higher values mainly due to greater reported consumption of milk and of vegetables. A 24-hour recall was as good as the FFQ in placing individuals in the distribution of habitual diet from weighed records. Results obtained from a 7-day estimated record were closest to those obtained from the weighed record. Correlations between 24-hour urine excretion and dietary N intake from weighed records were high (0.78–0.87) as were those with estimated food diaries (0.60–0.70). Correlations between urine N and the FFQ and 24-hour recall were lower (0.10 to 0.27), but improved by energy adjustment using residuals for N and K which are correlated with total energy intake. Comparisons between dietary estimates and urinary K and serum carotenoids and vitamin C showed broadly similar results. Limited biomarker information amongst 200 UK EPIC participants supported the findings of the validation study.

Conclusions. UK EPIC uses three methods (the 7-day diary, an improved FFQ, and the 24-hour recall) to assess diet. 93% of first food diaries are returned completed by participants. Repeated diaries are the main dietary assessment method for nested case-control analyses.

Keywords: dietary assessment, weighed food records, food records, 24-hour recalls, food frequency questionnaires, urine, nitrogen, potassium, serum carotenoids, EPIC

* MRC Dunn Clinical Nutrition Centre, Hills Road, Cambridge CB2 2DH, UK.
** Institute of Public Health, University of Cambridge, Forvie Site, Robinson Way, Cambridge CB2 2SR, UK.
† Clinical Gerontology Unit, F & G Block, Addenbrooke's Hospital, Cambridge, CB2 2QQ, UK.
‡ Human Nutrition Research Group, School of Biomedical Sciences, University of Ulster, Coleraine BT52 1SA, UK.
§ Imperial Cancer Research Fund, Cancer Epidemiology Unit, Gibson Building, The Radcliffe Infirmary, Oxford OX2 6HE, UK.
‖ Imperial Cancer Research Fund, General Practice Research Group, Gibson Building, The Radcliffe Infirmary, Oxford OX2 6HE, UK.

Plans for a UK prospective study of diet and cancer were underway before the collaboration between several centres of the EPIC (European Prospective Investigation of Cancer and Nutrition) was instigated. Hence the work to assess the validity of dietary methods to be used was begun at an earlier date than in other centres and with a validation protocol similar to that which had been suggested for the Malmo prospective study of diet and cancer.[1,2] This validation protocol had been devised from work conducted in a metabolic suite to assess the

utility of the 24-hour urine nitrogen technique to validate individual measures of dietary intake, and the development of a marker to verify the completeness of 24-hour urine collections.[3,4] This work showed that a minimum of 16 days of weighed records and eight 24-hour urine collections were required to assess usual dietary intake, and the validation protocol therefore specified 4 days of weighed food records and two 24-hour urine collections, to be repeated at 3-month intervals over a year. A set of dietary balances, called PETRA (Portable Electronic Tape Recorded Automatic) scales were also developed to minimize the burden on volunteers when asked to weigh and record their food for prolonged periods of time. PETRA is of additional benefit in dietary validation studies because weights of foods are recorded on tape and not disclosed to the participant.[1]

In the UK, the validity of seven different methods of dietary assessment suitable for the proposed cohort of 50 000 subjects was tested against the baseline validation protocol. The cohort was planned initially to be recruited from breast screening clinics and so women aged 50 to 65 years were chosen as the study group. The aim was to find a method that was able to assess usual diet and yet be suitable for large numbers of participants. The chosen method had also to cover intake of all items of diet since most items of diet have been linked to initiation, promotion, and prevention of cancer, and be flexible enough to cope with changes in hypotheses as the cohort progressed over time. The validity of the three methods finally chosen for use in the cohort is described here. These methods and details of the other four methods tested have also been described elsewhere.[5,6]

METHODS

Volunteers

All women aged 50–65 years from the lists of two general practices in Cambridge were contacted by post either in October 1988 (Group 1, first practice) or October 1989 (Group 2, second practice), and asked to fill in a general health, food and lifestyle questionnaire. Of the 1064 approached, 25% who answered 'Yes' to the question 'Would you be interested in more detailed studies of diet?' were contacted by telephone and the purpose of the study was explained. Of those 18% remained interested and were then visited at home for further discussion, and those who still wished to participate were entered into the study. No exclusions were made on grounds of ill health and the protocol was approved by the Dunn Nutrition Unit Ethics Committee in October 1988.

Protocol

Each individual was studied at home on four occasions (seasons) over the course of 1 year, and at each season was asked to complete 4 days of weighed food records. Season 1 was from October–January; season 2, February–March; season 3, April–June; season 4, July–September. The volunteers were also asked to provide two 24-hour urine collections on each occasion, so that over the year each individual provided 16 days of weighed dietary records, and eight 24-hour urine collections. Overnight fasting blood samples were also obtained, together with body weight, fasting breath samples, and information on vitamin supplement consumption at each occasion. Seven other dietary methods were investigated over the course of one year and of these, one food frequency questionnaire (FFQ), one 24-hour recall, and one estimated record (called a 7-day food diary) were chosen for the study method.

Weighed Records

The weighed records were obtained using PETRA scales (Cherlyn Electronics Limited, Cambridge). These are accurate to ± 1 g and automatically record verbal descriptions and weights on food on a dual track cassette, thus avoiding the necessity for subjects to keep written records. The PETRA scales do not disclose the weights of foods eaten to the subject.[1]

Subjects were visited in their homes the day before they were due to begin to weigh their food (Day 0) when they were given a demonstration of the PETRA scales and asked to try them out themselves. The following day (Day 1) they were revisited and the verbal descriptions recorded on the tapes were checked for completeness using a personal cassette player. Subjects were left with written instructions and with a notebook for recording recipes and food eaten out of the home which had not been recorded on the PETRA scales. The scales, notebook and completed tapes were collected from the subjects 1 or 2 days after completion of the 4-day weighing period. Each 4-day period included different days chosen to ensure that all days of the week were studied, and that there was an appropriate ratio of weekend to weekdays included during the year. Subjects who for technical and other reasons submitted unsatisfactory records for all or part of the 4 days were asked to weigh their food for extra days the following season if possible.

The weights and verbal descriptions of food consumed were transposed by hand from the tape cassettes using the PETRA master console, and hand-coded for computer calculation of nutrient intake using food tables.[7–9] Values for separate carotenoids are not included in this data base, and the values are mostly for

β-carotene. Subjects from practice 1 (Group 1) were studied from October–September 1988–1989, and those from practice 2 (Group 2) from October–September 1989–1990.

24-hour Urine Collections

Subjects were instructed on the technique of 24-hour urine collections on the first day of weighing their food, and asked to make the first collection on the third or fourth day. They were given a canvas bag containing two 2 l containers, each with 2 g boric acid as preservative, a jug, funnel, safety pin to pin to their underclothes as a reminder, and a set of instructions. They were asked to discard the first specimen of the day, for example at 7 a.m. and from then on to collect all specimens for 24 hours up to and including 7 a.m. the next day. They were given three 80 mg tablets of p-amino benzoic acid (PABAcheck, Laboratories for Applied Biology, London) to take with meals as a marker for completeness of 24 hour urine collections, [3] and asked to fill in a questionnaire with details of any medications, the time of the beginning and end of the collections, and whether or not they had any problems or missed specimens. The collection was returned with the weighing equipment on the day after completing the weighed records. At the same time a further set of instructions and equipment was given for the second 24-hour urine collection, to be made usually 3–4 days later. On return to the laboratory, the volume of the 24-hour collections was recorded, and aliquots stored at –20°C prior to analysis. Total urinary nitrogen was determined by the Kjeldahl technique (Tecator 1002, Perstorp Analytical, Bristol) and sodium and potassium by flame photometry. PABA was measured colorimetrically.[10]

Fasting Blood Collection and Anthropometry

Subjects were visited at home by the study nurse within 7 days of completing each 4-day diet record by prior arrangement between 6 a.m. and 9 a.m. after overnight fasting and before breakfast. The second 24-hour urine collection was picked up, and subjects were weighed on a spring balance without shoes in light clothing. Height was also measured without shoes using a Nivotoise portable height measure (CMS Equipment, London). An estimate of Basal Metabolic Rate (BMR) was calculated from the equations of Schofield *et al.*[11]

A 30 ml sample of blood was taken by venepuncture, 20 ml in lithium heparin, and 10 ml without anticoagulant. The samples were returned to the laboratory within 3–4 hours in an insulated picnic box, and centrifuged at 3000 r.p.m. for 10 mins at 10°C. 0.2 ml plasma was added to 0.4 ml 10% metaphosphoric acid and stored at −40°C prior to analysis for vitamin C. Remaining plasma was stored at −40°C prior to analysis for carotenoids. Plasma vitamin C was measured by liquid chromatography with electrochemical detection in both groups.[12,13] Due to technical problems and storage effects, results for calculating means were only available from seasons 3 and 4 in group 1, and from seasons 1 and 2 in group 2. In group 1 only, carotenes, retinol and tocopherols were measured using absorptiometric detection.[14] The intra-batch precision of these methods is 5–10% coefficient of variation.

Food Frequency Questionnaire

The food frequency questionnaire (FFQ) was based on that used in the US Nurses' Health Study.[15,16] The frequency categories were not changed, but the lists of foods were modified by changing American food names to their British equivalent and by using the National Food Survey to identify additional foods which are important sources of nutrients in average British diets.[17]

Nutrient intakes were calculated by multiplying the frequency of food consumption by standard portion weights to obtain grams of each food consumed per day; these were then converted to nutrient intake using an appropriate food table code. 'Medium serving' or units were specified (pints, slices, teaspoons, etc.). The weights of 'medium serving' portions were derived from experience with other dietary surveys and from published values.[18] Questionnaires were excluded if 10 or more lines had not been filled in, or if a frequency of four or more per day of designated items (for example cabbage) had been chosen. The questionnaire was given to subjects to complete immediately before they started to weigh their food on day 0 in season 3.

24-hour Recall

The 24-hour recall was self completed and unstructured and consisted of a blank sheet of paper and written example with portions in household measures and was given to the subjects during recruitment before the start of season 1. Subjects' descriptions of units and published average portion weights were used to calculate nutrient intakes.[18] Data were hand-coded for computer calculation of nutrient intakes, using the same food tables as were used for the weighed records.

Estimated Diet Records

These were left with the subjects on day 5 or 6 of the protocol to fill in within the next 14 days, with a stamped addressed envelope for postal return, in group 2, season 3 only. The record was an open-ended (unstructured) estimated diet record. This 7-day food

diary had been developed for the MRC National Survey of Health and Development.[19,20] Subjects kept a written record of food consumed at the time of eating. The booklet contained 15 sets of black and white photographs, taken mostly from the study by Edington *et al*. in 1989.[21] Each set of photographs had three portion choices. Subjects were allowed to state portion sizes in other measures if they so wished. A computer program, DIDO (Data In, Diet Out) was developed specifically for the coding of these booklets for nutrient analysis.[20] For this diary only, the nutrient composition database was different from that used in the weighed records and the other methods. It included values from the food table supplements for Cereals and for Milk and Milk Products.[22,23]

Methods Used in the UK Norfolk EPIC Cohort

Three methods are used to assess diet in EPIC UK, a 24-hour recall, the 7-day diary, and the FFQ, modified to improve estimates of milk and breakfast cereal consumption. Men and women aged 45–75 are recruited from the lists of general practitioners in Norfolk. With the letter of introduction and health and lifestyle questionnaire, they are sent the unstructured 24-hour recall which is returned completed if they agree to participate. The FFQ is sent out with an appointment to see the EPIC nurse, when blood is taken, and anthropometry and respirometry is carried out. The FFQ is checked for completeness by the nurse, who then goes on to explain the procedure for completing the food diary, using the previous day's food intake as an example. The procedure takes approximately 15 minutes, and a carbon copy is retained of this record. The participants are then asked to fill in the remaining 6 days of the diary record and to return the completed diary in a pre-paid envelope. On average 93% of these completed diaries are returned to EPIC. Preliminary results from these methods obtained from an age-stratified random sample of the first 2000 recruits are presented here, in comparison with serum carotenoids and vitamin C from the single blood specimen collected from participants in the main study.[24]

Statistics

Means and standard deviations of untransformed data are presented, using the averages of all available days of weighed records and 7 days of food diary for each individual. The significance of the differences were assessed using paired t-tests, except where otherwise stated. Using SAS (version 6.07), distributions were ranked and then divided into four equal parts and results were tabulated for the weighed records and other methods, in order to see if individuals were ranked similarly by different methods. In Tables 1 and 2, results from only those 146 individuals who had completed the weighed record exactly, that is who submitted four complete dietary records at each of the four seasons, were used for this and Spearman rank correlation analysis between methods. Results are given for energy, macronutrients, non-starch polysaccharides (NSP), potassium, calcium, iron, carotene, retinol, and vitamin C, all of which had less than 1% 'missing values' in the data base. Results are also shown for 'dietary fibre', although the data base contained 9% missing values.

All available weighed intake data were used for correlations with biological variables. To compare dietary intakes of vitamin C with and without supplements, the average amount of vitamin C calculated to have been taken from supplements was added on to results from each method. Separate analyses were also conducted in those who did not take vitamin C supplements.

RESULTS

Participants

In group 1 86 subjects and in group 2 96 subjects returned completed questionnaires and undertook to take part in the study in the first season. Of these, 79 in group 1 and 81 in group 2 completed most or all of the protocol over the year, about 15% of the original sample. There were no significant differences between the two groups in average age, weight, or height. There were also no significant differences between the two groups in mean energy, macronutrients, NSP or micronutrient intakes calculated from the weighed records. Except where otherwise stated, the data were therefore combined for the two groups, giving a total of 160 subjects, who completed the study. There was no significant difference in body weight between the group of 160 women studied here and a representative British population sample of 283 women,[25] but the Cambridge women were significantly taller, and reported significantly greater intakes of energy, macronutrients, vitamin C, iron and calcium, and had lower levels of serum tocopherol but higher levels of serum carotenoids.[5,6] A total of 160 subjects completed seasons 1–3, but four subjects had moved by season 4, so that 156 completed all four seasons. The results of the four subjects who completed only three seasons of data are included in the biomarker comparisons.

Comparisons between Weighed Records, the FFQ, 24-hour Recall and Food Diary

Of the FFQ 33 were excluded due to missing information. Table 1 shows that the questionnaire significantly

TABLE 1 *Means and standard deviations for daily intakes of nutrients obtained from 16-day weighed records completed by 160 women, and 3 different dietary methods. Spearman correlation coefficients between individual results from weighed records and food frequency questionnaires completed by 127 women, 24-hour recalls completed by 146 women, and 7-day estimated diet records completed by 73 women are also shown*

	16-day weighed records		FFQ			24-hour recall			7-day estimated food record (food diary)		
	Mean	SD	Mean	SD	r	Mean	SD	r	Mean	SD	r
Energy MJ	7.83	1.50	***8.81	2.40	0.52	7.57	2.2	0.42	7.89	1.49	0.59
Fat g	76	19	***87	31	0.55	77	34	0.40	*80	23	0.63
Protein g	69	12	***82	22	0.43	70	24	0.21	69	12	0.66
Carbohydrates g	223	52	**245	77	0.55	*206	68	0.60	215	47	0.71
Sugars g	109	35	***135	55	0.51	102	42	0.63	106	33	0.77
Starch g	108	27	***93	33	0.53	*98	41	0.56	104	25	0.70
NSP g	16	5	***19	7	0.57	15	6	0.61	*15	4	0.74
'Fibre' g	22	7	***27	10	0.55	21	10	0.58	22	6	0.73
Potassium g	3.2	0.6	***4.0	2.2	0.39	3.1	0.9	0.56	3.2	0.6	0.76
Calcium mg	952	245	***1308	453	0.50	879	360	0.28	*864	248	0.67
Iron mg	12.9	3.8	13.6	7.0	0.43	12.3	5.9	0.53	12.4	2.9	0.83
Carotene mg	3.4	1.9	***5.1	3.2	0.45	3.5	3.7	0.28	3.2	1.8	0.66
Retinol μg	797	773	1010	952	0.55	1070	4178	0.54	740	821	0.35
Vitamin C mg	99	41	122	53	0.54	**87	56	0.54	106	48	0.70
Alcohol g	9	13	8	11	0.90	**7	14	0.60	9	10	0.88
Fat % energy	35.9	4.6	36.3	6.1	0.64	36.5	8.9	0.49	*37.0	5.7	0.77
Protein % energy	15.2	2.3	***16.1	2.7	0.70	16.5	4.9	0.45	15.2	2.7	0.81
Carbohydrate % energy	45.6	5.8	44.6	6.5	0.69	*44.0	9.0	0.57	***43.8	5.7	0.81

* $P < 0.01$ Significantly different from weighed records.
** $P < 0.001$ Significantly different from weighed records.
*** $P < 0.0001$ Significantly different from weighed records.
95% confidence intervals for n = 150 are –0.06–0.26 for r = 0.1; –0.04–0.36 for r = 0.2; 0.16–0.44 for r = 0.3; 0.26–0.54 for r = 0.4; 0.38–0.62 for r = 0.5; 0.50–0.70 for r = 0.6; 0.68–0.78 for r = 0.7; 0.74–0.86 for r = 0.8; 0.86–0.94 for r = 0.9.

overestimated most nutrients when compared with weighed records. The differences in carotene, vitamin C and NSP were partly accounted for by the approximately 120 g daily greater reported consumption of vegetables in the FFQ, due to a greater reported frequency of consumption than measured by the weighed records.[5] Other overestimates of intake by FFQ were: daily milk consumption (150 g greater), cheese (15 g), and coffee (160 g), which largely accounted for the significant differences in energy, fat, protein, potassium, calcium and sugars found between the FFQ and weighed records.[5]

Table 1 shows that, apart from vitamin C, starch, carbohydrates, and alcohol, there were no other differences between the weighed records and the 24-hour recall in reported average intakes of nutrients. Seasonal variations contributed to the significantly lower estimate of vitamin C consumption by the 24-hour recall method, since there was no significant difference between vitamin C assessed by the 24-hour recall and by the 4 days of weighed records collected at season 1, the season closest to the collection of the 24-hour recall.[5]

There were few differences in mean intakes of nutrients measured by the food diary method compared with those measured by the 16 days of weighed records, Table 1. The differences in fat, NSP and calcium were no longer significant when compared with the averages of nutrients assessed at the same season the diary was administered, season 3.[5] There were no significant differences in daily average food consumption between the two methods.

Table 1 also shows Spearman rank correlation coefficients between values obtained from each method and the weighed records. Correlation coefficients were highest for alcohol, and lowest for protein and carotene. Correlations with the FFQ ranged from 0.39 to 0.57 and with the 24-hour recall from 0.21 to 0.63, excluding alcohol. Correlations were of a greater magnitude between the 16 days of weighed records and the 7-day open-ended food diary, 0.35 to 0.83 excluding alcohol. Energy adjustment improved the magnitude of correlation coefficients between methods, but did not alter the ranking of each method compared with weighed records.[26,27]

TABLE 2 *Comparison of the % classification of nutrients from the food frequency questionnaire (FFQ) into the same and extreme quartiles of the distribution of nutrient intake as that obtained from the mean of 16-day weighed records completed by 127 women, from 24-hour recalls completed by 146 women, and from 7-day estimated diet records completed by 73 women*

	FFQ		24-hour recalls		7-day estimated food record	
	Same quartile	Extreme quartile	Same quartile	Extreme quartile	Same quartile	Extreme quartile
Energy	46	2	37	6	40	4
Fat	38	2	38	6	44	3
Protein	39	4	34	9	49	0
Carbohydrates	39	5	41	1	38	0
Sugars	44	5	56	1	58	1
Starch	43	5	46	3	47	0
NSP	43	2	44	3	51	1
'Fibre'	47	2	49	3	47	0
Potassium	35	6	42	1	58	1
Calcium	46	6	35	7	51	1
Iron	39	4	41	1	56	0
Carotene	40	6	30	7	42	0
Retinol	47	3	45	1	37	10
Vitamin C	37	2	39	3	48	0
Alcohol	75	0	39	9	70	0

Table 2 shows the extent to which each method was able to classify individuals into the same quartile of intake and the amount of misclassification into opposite quartiles compared with the weighed records. Alcohol was poorly classified by the 24-hour recall but for other methods it was correctly classified in over 70% of individuals and no individuals were misclassified into the extreme quartile. In general, all three methods correctly classified 30–50% of individuals into their correct quartile of intake for most nutrients, and misclassified in extreme quartiles 0–10%. Overall the 7-day estimated record classified the fewest individuals into the incorrect quarter of the distribution for most nutrients, although it was less accurate than other methods for retinol.

24-hour Urine Collections

In 16 of the 79 subjects in group 1 and 22 of 81 subjects in group 2 all eight 24-hour collections contained more than 85% of the PABA marker, and were designated complete. Completeness could not be assessed in all eight of one subject's collections, and in a further four isolated samples, due to interference from a medication containing paracetamol. These samples were not included in the analysis. Three subjects submitted only one complete collection, five subjects two, and eight subjects three collections that were designated complete. The remaining 105 subjects submitted between four and seven complete collections. There were highly significant differences in total nitrogen, potassium, sodium, and volume between the samples designated complete and incomplete by the PABA technique[6] and only those that were complete were used in further analysis.

Comparison of 24-hour Urine N Output with Recorded N Intake from Weighed Records and Grouping into Quintiles

To assess the validity of the weighed records, average dietary N intake from the 16 weighed records was compared with the 24-hour urine N output from all 24-hour urine collections.[3] Comparisons between urine and dietary nitrogen intake were not possible for the three individuals who submitted only a single complete urine collection, and the individual who had been taking paracetamol medication. In the remaining 156 individuals, average N intake from the 16-day records was 11.2 ± 2.3 g per day, and that from N excretion in the complete 24-hour urine 9.84 ± 1.78 g per day, so that the average ratio of urine N to dietary N was 0.91 ± 0.09. This was rather greater than the ratio of 0.81 ± 0.05 expected if the average results from all individuals were valid.[4]

To determine which, if any, of the individual results were valid, the distribution of the ratio of urine to dietary N was examined. The distribution of the ratio was not normal and coefficients of skewness and kurtosis were 1.21 and 2.00. In the absence of a clear

TABLE 3 *Means, SE, and correlation coefficients of anthropometric and other data from 156 individuals grouped into quintiles of the distribution according to their urine nitrogen to dietary nitrogen ratio from 16 days weighed records*

	Lower quintile n = 31		2nd quintile n = 31		3rd quintile n = 32		4th quintile n = 31		Top quintile n = 31	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
UN/DN ratio[a]	0.76*	0.005	0.83*	0.003	0.89*	0.003	0.96*	0.004	1.13*	0.022
Range	0.69–0.79		0.79–0.86		0.86–0.93		0.93–1.00		1.00–1.47	
Urine N g	8.76	0.23	9.74	0.36	10.34	0.27	10.56	0.35	10.89	0.30
Dietary N g	11.55	0.29	11.78	0.43	11.25	0.30	11.00	0.36	9.69	0.28
r UN v DN	0.976		0.994		0.989		0.994		0.778	
Intake (EI)[b] MJ	8.35	0.17	8.84	0.30	7.87	0.18	7.50	0.22	6.65′	0.23
EI/BMR ratio[c]	1.54	0.03	1.60	0.05	1.40	0.03	1.32	0.04	1.15	0.04
r UN/DN v EI/BMR	0.20		–0.13		0.07		0.03		–0.62	
Urine K mmol	65.55	2.11	74.06	3.20	75.09	3.25	72.97	3.53	75.37	4.09
Diet K mmol	82.45	2.15	88.59	3.67	84.36	3.08	77.67	3.31	73.82	2.97
UK/DK ratio	0.798	0.016	0.848	0.023	0.887	0.021	0.942	0.020	1.025	0.037
r UK v DK	0.768		0.903		0.799		0.888		0.740	
Weight kg	61.54	1.44	63.44	1.69	65.76	1.33	65.65	1.58	71.99′	2.67
Height m	1.60	0.02	1.62	0.01	1.65	0.01	1.65	0.01	1.64	0.01
BMI[d] kg/M^2	23.48	0.58	23.88	0.65	23.94	0.50	24.40	0.50	26.59′	0.97
Kg change over year	0.37	0.35	0.72	0.51	–0.27	0.40	1.18	0.40	0.02	0.45
Serum cholesterol mmol/L	5.81	0.18	6.20	0.18	6.00	0.23	6.03	0.19	5.69	0.20
Plasma vitamins µmol/l (for items other than vit C, data is for group 1)										
Vitamin C	69	5	59	4	62	4	60	4	61	5
Retinol	2.0	0.1	2.0	3.8	2.0	0.1	2.0	0.1	2.0	0.1
a tocopherol	27	2	26	1	26	1	28	2	26	1
g tocopherol	1.9	0.2	2.4	0.3	2.1	1.2	2.4	0.2	2.4	0.3

* $P < 0.001$ between all quintiles, ′ $P < 0.05$ between top and 4th quintile, ″ $P < 0.05$ between top and bottom quintile.
[a] UN/DN ratio = urine nitrogen/dietary nitrogen ratio.
[b] EI = energy intake.
[c] BMR = basal metabolic rate.
[d] BMI = body mass index.

bimodal distribution, results were sorted and examined as quintiles of the distribution of the urine to dietary N ratio. Means of this ratio varied from 0.76 in the lower quintile of the distribution to 1.13 in the upper quintile (Table 3). Correlations between urine and dietary N were 0.98 to 0.99 in the bottom four quintiles of the distribution, but lower (0.78) in the top quintile.

Energy intakes, calculated BMR and the calculated ratio of energy intake to BMR are also shown in Table 3. The average ratio of energy intake to BMR of 1.15 in the top quintile was significantly different ($P < 0.05$) from that of the fourth quintile, but there was no significant difference in means between the fourth and third quintile ($P < 0.05$). The ratio of energy intake to BMR was not correlated with the urinary to dietary N ratio in the lower four quintiles of the distribution (r = 0.20 to 0.12). However, there was a significant inverse correlation in the top quintile (r = –0.62) so that most individuals with an EI:BMR ratio of less than 1.2 were also classified into the top quintile. Overall 36 (23%) of the 159 individuals had an EI:BMR ratio of less than 1.2, compared with 20% classified into the top quintile by the urine to dietary N ratio. The 13 subjects who submitted only two or three complete collections were fairly evenly distributed throughout the quintiles of the distribution and their ratios of energy intake to BMR showed the same trends as the complete quintiles, with average ratios of 1.39, 1.58, 1.48, 1.23, and 0.98 (data not shown).

Table 3 shows anthropometric and other data from 156 individuals grouped into quintiles. There were clear trends in the ratio of urine potassium to dietary potassium associated with the quintiles of urine to dietary N ratio and correlations of 0.74 to 0.90 between urine and dietary potassium within quintiles. Individuals in the top quintile had a significantly greater

body mass index (BMI) and weight than those in the fourth quintile ($P < 0.05$), but there were no significant differences between the average BMI and weight of individuals in the fourth compared with those in the third quintile ($P < 0.05$). Individuals in the lower, second and fourth quintile put on 0.4 to 1.2 kg over the year, but those in the top quintile did not gain or lose weight and those in the third quintile lost 0.3 kg weight. There was no consistent trend with height, although individuals in the lower quintiles of the distribution were 2 and 5 cm shorter on average than in the other quintiles.

Differences in Food and Nutrient Intakes by Top Versus Other Four Quintiles of the Ratio of Urine to Dietary N

The data shown in Table 3 indicated that mean values from individuals assigned to the top quintile were different from the others. Data for both weighed records and other methods were therefore recalculated and considered separately for individuals in the top quintile and for individuals in the other four quintiles of the distribution in urine to dietary N ratio, who can be considered to have recorded valid estimates of usual diet. Intakes of energy and all energy yielding nutrients calculated from weighed records in individuals in the top quintile were significantly lower than those from individuals in the other quintiles (Table 4). On average there was an 18 g difference in reported fat consumption, and a 27 g difference in reported sugars consumption between the average values reported in the top and the other four quintiles according to the urine to dietary N ratio. Mean consumption of cakes, breakfast cereals, milk, eggs, fats, and sugars was also significantly lower in those individuals classified in the top quintile of the distribution (data not shown).

Using the FFQ, individuals in the top quintile reported significantly lower mean intakes of energy, protein, starch, sugars, calcium and potassium intakes than those in the other quintiles (Table 4). However, apart from a lower intake of sugars (106 ± 4 g lower quintiles, 87 ± 7 g top quintile, $P < 0.01$), there were no significant differences between quintiles with results obtained from the 24-hour recall, and there were no differences at all between quintiles when results from the estimated food record (7-day diary) were compared.[6]

TABLE 4 *Means (SE) of daily intake of nutrients from 156 individuals grouped into top versus the other four quintiles of the distribution according to their urine to dietary nitrogen ratio from 16-day weighed records, and from the same individuals, but using results from the food frequency questionnaire*

	Lower four quintiles n = 125	Top quintile n = 31	*p* top versus lower four quintiles
Weighed records			
Energy MJ	8.14 (0.12)	6.65 (0.23)	<0.001
Protein g	71 (1)	60 (2)	<0.001
Fat g	80 (2)	62 (2)	<0.001
Starch g	113 (3)	100 (5)	<0.05
Sugars g	115 (3)	88 (5)	<0.001
Calcium mg	997 (21)	781 (35)	<0.001
Vitamin C mg	100 (3)	94 (8)	ns
NSP g	16.1 (0.5)	14.6 (0.8)	ns
Alcohol	9 (1)	8 (2)	ns
Food frequency questionnaires			
	n = 114	n = 22	
Energy MJ	9.07 (0.23)	7.56 (0.37)	<0.01
Protein g	84 (2)	74 (3)	<0.01
Fat g	90 (3)	74 (4)	ns
Starch g	103 (3)	87 (6)	<0.02
Sugars g	140 (5)	113 (8)	<0.01
Calcium mg	1370 (20)	1033 (58)	<0.001
NSP g	19.6 (0.7)	18.6 (1.2)	ns
Alcohol g	8.5 (1.0)	7.3 (2)	ns
Vitamin C mg	123 (2)	121 (11)	ns

Comparisons between Assessments from Different Methods and Biological Indices of Diet

The validity of dietary methods was considered by comparisons with the biological markers 24-hour urine N and 24-hour urine K, in the 156 individuals as a whole, in the 80% individuals who had submitted valid records as judged by the urine to dietary N ratio, and in the 20% who had not. Table 5 shows means and correlation coefficients between intake values of N, and potassium from the weighed records, the FFQ, the 24-hour recall and the 7-day diet diary. Correlations between dietary N intake and dietary assessment methods were highest for the weighed records, 0.87 lowest four quintiles, 0.78 top quintile, 0.69 all values combined. Correlations between urine and dietary N from the FFQ and the 24-hour recall were much lower, 0.10 and 0.24, and results from the diary were closest to those obtained with weighed records, correlation 0.65 for all individuals. There did not appear to be differences between groups of individuals classified into the top versus other four quintiles, apart from a negative correlation (–0.31) between urine and dietary N in the top quintile of 24-hour recall data and a positive correlation (0.50) in the top quintile of the FFQ.

Correlations between potassium intake estimated from the weighed records, food diary, and 24-hour recall showed the same general pattern as those found with 24-hour urine N. Correlations were highest

TABLE 5 *Comparisons between dietary nitrogen and potassium intake from different methods of dietary assessment with 24-hour urine nitrogen and potassium in 156 individuals grouped into upper versus lower four quintiles of the distribution according to their ratio of urine nitrogen to dietary nitrogen from 16-day weighed records*

	1–4th quintiles	SE	Top quintile	SE	*P* top versus 1–4th quintiles
Averages of N intake (g per day) from:					
16-day weighed records	11.4	0.17	9.7	0.28	$P < 0.001$
FFQ[a]	13.5	0.35	11.7	0.51	$P < 0.01$
24-hour recall	11.2	0.27	11.9	1.07	ns
Estimated food record[a] (7-day diary)	11.2	0.25	10.8	0.55	ns
Averages of K intake (mmol per day) from:					
16-day weighed records	83	1.5	74	2.9	$P < 0.05$
FFQ[a]	104	2.6	93	3.9	$P < 0.05$
24-hour recall	80	2.1	77	4.1	ns
Estimated food record[a] (7-day diary)	81	2.0	82	3.6	ns

	1–4th quintile	Top quintile	All values	Energy adjusted All values
Correlations between 24-hour urine (N)-versus:				
N from 16-day weighed record	0.87	0.78	0.69	0.69
N from FFQ	0.27	0.50	0.24	0.49
N from 24-hour recall	0.26	–0.31	0.10	0.09
N from food record (7-day diary)[b]	0.70	0.60	0.65	0.67
Correlations between 24-hour urine (K)-versus:				
K from 16-day weighed record	0.82	0.74	0.76	0.76
K from FFQ	0.28	0.28	0.25	0.41
K from 24-hour recall	0.52	0.53	0.51	0.54
K from food record (7-day diary)	0.70	0.57	0.66	0.64

[a] n = 136 for Food Frequency Questionnaire (FFQ), 63 for 7-day diary.

[b] Correlations for urine and dietary N for women of group 2 (n = 81) and weighed records were 0.88, 0.94 and 0.79 in quintiles 1–4, top quintiles, and all values respectively.

95% confidence intervals for n = 150 are –0.06–0.26 for r = 0.1; –0.04–0.36 for r = 0.2; 0.16–0.44 for r = 0.3; 0.26–0.54 for r = 0.4; 0.38–0.62 for r = 0.5; 0.50–0.70 for r = 0.6; 0.68–0.78 for r = 0.7; 0.74–0.86 for r = 0.8; 0.86–0.94 for r = 0.9.

(0.74–0.82) for the weighed records, next best for the food diary (0.53–0.70) and much lower (0.25–0.28) for the FFQ and 24-hour recall (0.51–0.53), Table 5.

Table 5 also shows correlation coefficients between urine biomarkers and intakes adjusted for energy using the residual method.[26–28] There were strong positive correlations between energy intake and unadjusted N intake,[27] and results from the FFQ were improved when compared with biomarkers, for example, the correlation between dietary and urinary N increased from 0.24 unadjusted to 0.49 adjusted (Table 5). To examine the effect of temporal relationships of the biomarkers and the different dietary methods, mean N intake from the last two seasons (8 days) of weighed records, and the other three methods was compared with urine N excretion from the very first 24-hour urine collection in season 1. Correlations between N from this single 24-hour urine and dietary N were 0.50 for the weighed records, 0.54 for the estimated record, 0.25 for the FFQ, and 0.05 for the 24-hour recall. In the 69 individuals who completed all four different methods, the correlation between urine N and dietary N using weighed records was 0.83, using the diary 0.67, using the FFQ 0.30, and using the 24-hour recall 0.12.

Estimates of carotene, mainly β-carotene, only were available in the database to calculate carotene intake. There was no effect of underreporting on carotene intake but there were highly significant differences

TABLE 6 *Comparisons between carotene intake from different methods of dietary assessment with plasma carotenoids in 156 individuals*

Grouped into upper versus lower four quintiles of the distribution according to the ratio of urine nitrogen to dietary nitrogen from 16-day weighed records

	1–4th quintiles	SE	Top quintile	SE	*P* top versus 1–4th quintiles
Averages of intake from 16-day weighed records					
Carotene (mg per day)	3.50	0.17	3.54	0.37	$P > 0.05$
Vitamin C (mg per day)	100	3.5	94	7.7	$P > 0.05$
Vitamin C (mg per day), with supplements (medians)	101		97		
Plasma carotenoids and vitamin C μmol/l					
Lutein	0.44	0.02	0.32	0.03	<0.001
β-cryptoxanthin	0.28	0.02	0.24	0.04	$P > 0.05$
Lycopene	0.32	0.02	0.24	0.03	<0.05
α-carotene	0.12	0.01	0.07	0.01	<0.001
β-carotene	0.55	0.03	0.35	0.04	<0.001
cis carotene	0.05	0.01	0.04	0.01	$P > 0.05$
Vitamin C	62	2.1	61	5.1	$P > 0.05$

Correlations between dietary carotene from different dietary methods and plasma carotenoids (Group 1)

	Weighed records	FFQ	24-hour recall	7-day checklist
Lutein	0.36	0.03	0.05	0.21
β-cryptoxanthin	0.20	–0.02	–0.03	0.07
Lycopene	0.33	0.17	0.08	0.21
α-carotene	0.62	0.42	0.19	0.34
cis carotene	0.35	0.14	0.00	0.13
β-carotene	0.48	0.15	0.08	0.28

Correlations between dietary vitamin C from different dietary methods and plasma vitamin C seasons 3 and 4 (group 1) and seasons 1 and 2 (group 2). Spearman rank correlation coefficients

	Weighed records	FFQ	24-hour recall	7-day diary
Excluding supplements (n = 156)	0.47	0.21	0.30	0.22
Including supplements (n = 156)	0.51	0.35	0.34	0.22
Non-supplement users (n = 127)	0.49	0.26	0.26	0.22

95% confidence intervals for n = 150 are –0.06–0.26 for r = 0.1; –0.04–0.36 for r = 0.2; 0.16–0.44 for r = 0.3; 0.26–0.54 for r = 0.4; 0.38–0.62 for r = 0.5; 0.50–0.70 for r = 0.6; 0.68–0.78 for r = 0.7; 0.74–0.86 for r = 0.8; 0.86–0.94 for r = 0.9.

between the top quintile and other quintiles in plasma lutein, β- and α-carotene values (Table 6). Correlations between BMI and plasma carotenoids were somewhat higher than those between the urine to dietary N ratio. For example, the correlation with plasma β-carotene and urine to dietary N ratio was –0.33, whereas that with BMI was –0.46.

The correlations between dietary estimates from different methods and plasma carotenoids are shown for the group as a whole in Table 6. The correlation between diet estimates of carotene from weighed records and plasma concentrations of β-carotene was 0.48, but for the other carotenoids ranged from 0.62 for plasma α-carotene, to 0.20 for plasma β-cryptoxanthin. Correlations between serum carotenoids and dietary intakes were highest using weighed records; the correlation between plasma β-carotene and dietary carotene from weighed records was 0.48, but 0.15 and 0.08 using values for dietary carotene from the FFQ and 24-hour recall. Plasma carotenoids were not analysed in group 2,

TABLE 7 *Comparison of results obtained by three dietary methods from a random sample of the first 200 individuals recruited into the UK EPIC cohort*

Differences in mean intakes between methods (*$P < 0.05$ v. diary)

Nutrient	Diary		FFQ		24-hour recall	
	Mean	SD	Mean	SD	Mean	SD
Energy MJ	8.67	2.56	8.81	2.72	8.72	3.07
Protein g	80.3	21.7	82.3	20.9	*87.8	31.4
Fat g	82.7	30.6	80.9	33.9	83.0	41.4
Carbohydrate g	250	81	*262	86	250	94
Sugars g	111	45	*135	55	109	52
NSP g	16	6	*19	7	16	7
Vitamin C mg	77	42	*114	61	*87	62
β-carotene mg	1801	1093	*2898	1567	*2229	2106
Calcium mg	907	326	*1044	313	*1069	495
Alcohol g	8	12	7	11	*5	12

Spearman correlation coefficients between plasma carotenoids and vitamin C, and β-carotene equivalents and vitamin C intakes

Plasma analytes	Diary	FFQ β-carotene equivalents	Recall
α-carotene	0.34	0.28	0.15
β-carotene	0.21	0.12	0.13
α-cryptoxanthin	0.10	–0.08	–0.04
β-cryptoxanthin	0.21	0.08	–0.02
Lutein	0.13	0.11	0.02
Lycopene	0.20	–0.01	–0.01
Vitamin C (all)	0.48	0.46	0.42
(no supplements, n = 164)	0.36	0.36	0.27

95% confidence intervals for n = 200 are –0.04–0.24 for r = 0.1; –0.06–0.34 for r = 0.2; 0.18–0.42 for r = 0.3; 0.28–0.52 for r = 0.4; 0.40–0.60 for r = 0.5; 0.50–0.70 for r = 0.6; 0.62–0.78 for r = 0.7; 0.74–0.86 for r = 0.8; 0.88–0.92 for r = 0.9.

so a direct comparison with the 7-day diary is not possible. However, correlations with carotene intake assessed from a similar method, a structured 7-day food checklist[5] are shown in Table 6. In general, correlations between dietary carotene intake using this method and plasma carotenoids were higher than those obtained with intakes derived from the 24-hour recall and FFQ. The correlation between plasma β-carotene and dietary carotene from this method for example was 0.28, lower than those with weighed records, but higher than those found with the FFQ and 24-hour recall.

There was also no effect of underreporting on estimates of intake of vitamin C from weighed records, and no systematic difference in plasma values of vitamin C in underreporters compared with others (Table 6). The correlation between the averages of the two plasma vitamin C and averages of dietary vitamin C from the 16-day weighed records were 0.47, and 0.51 including supplements. In non-supplement users, the rank correlation coefficient was similar, 0.49 (Table 6). Values obtained from correlation of plasma vitamin C with intakes estimated from the other methods were all lower than those obtained from the weighed records and of a similar order 0.2–0.3, higher values being obtained when supplement intakes were added on to the estimates of intake by FFQ and 24-hour recall (Table 6).

Analysis of Dietary Intake from the Subsample of the UK EPIC Cohort

Table 7 shows mean intakes of selected nutrients assessed by all three methods in the random sample of 200 EPIC participants. With the improved FFQ, differences between it and the 7-day diary were reduced, although intakes assessed by the FFQ remained significantly higher for sugars, NSP (non-starch polysaccharides), vitamin C and β-carotene equivalents. 24-hour

urine collections are not made in the EPIC cohort, so biomarker data to compare with the dietary data are limited to plasma values derived from a single non-fasting blood specimen. These preliminary data suggest that correlations with dietary intake data from the diary are more strongly correlated with plasma carotenoids and vitamin C than those obtained from the FFQ and 24-hour recall (Table 7). Vitamin C supplements, however, have a major effect on the correlation between intake estimates and plasma levels (Table 7).

DISCUSSION

FFQ are the most commonly used dietary assessment technique used in nutritional epidemiology, and are designed to assess habitual food and nutrient intake. 24-hour recalls are also commonly used but their limitations for individual assessments due to attenuation from daily variation in nutrient intake are well known.[1,29–31] It was therefore surprising to find that the FFQ was not appreciably better than the 24-hour recall at placing individuals in the distribution of habitual intake for the majority of nutrients when compared with weighed records of food intake (Tables 1, 2). High correlations were only obtained for alcohol consumption in the FFQ, but this is a common finding due to the fact that a substantial proportion of the individual values obtained are zero, leading to a wide range in intake, and that alcohol is generally consumed in standard units.

However, the FFQ used here does not appear to be less accurate than in other published comparisons of regularly used questionnaires of similar length, at least when compared with records of food intake. Alcohol apart, the correlation coefficients between nutrients assessed from the questionnaire and from weighed records were generally of the order of 0.4–0.6 (Table 1). Correlations are similar to values obtained elsewhere in comparative validation studies.[28–33]

Tables 1 and 2 show that both correlation coefficients and classification into quartiles gave similar results in comparing the precision of simpler methods of dietary assessment with weighed records. Using both criteria, the 7-day estimated diet record gave the best agreement with weighed records, followed by the 24-hour recall and FFQ. The low correlations with single 24-hour recalls are to be expected from the known effect of attenuation due to day to day variation. Correlations between different dietary methods tend to be lower in studies of selected women than in less selected samples and can be improved by energy adjustment.[34] However, in the present study, energy adjustment, although generally improving the magnitude of correlation coefficients, did not alter the ranking of the precision of different methods when compared with weighed records.[26,27]

Results from the FFQ also gave significantly higher values for most nutrients than the weighed records, 24-hour recall and food diary. This finding, that FFQ results yield higher average daily intakes, particularly of vegetables, than other methods has been noted before.[31,33,35,36] In the present study, differences were largely attributed to a greater reported frequency of consumption of vegetables by participants using the FFQ than reported on the weighed records, and probably to an inappropriately large reported portion size for milk in particular.[5] Errors from the use of average portion sizes, derived from both male and female populations[18] were used for all simpler methods of dietary assessment tested here and hence are unlikely to be a particular problem with the FFQ.

Comparisons between methods are of limited use in estimating validity, since all methods of dietary assessment are subject to errors.[1] To examine which if any method gave valid estimates of usual diet, comparisons were made between the intake values obtained by the different methods and the biological markers of food intake in 24-hour urine collections and plasma. Only 24-hour urine collections shown to be complete by the PABAcheck method were used to assess validity since those that were incomplete contained systematically less total nitrogen and potassium.[6] Comparisons with the urine to dietary N ratio and other differences shown in Table 3 demonstrated that individuals in the top quintile were not reporting their usual diet. Furthermore, these individuals were as likely to underreport compared with others using the FFQ as they were when asked to keep weighed records (Table 4). The average ratio of energy intake to BMR from weighed records kept by these individuals was 1.15, below the physiological limits that would match energy expenditure with no gains or losses of weight.[37] Since, over the year, this group neither gained nor lost weight, it has to be concluded that individuals in this 20% of the distribution did not report valid estimates of their usual diet. This group were heavier and had significantly lower plasma values of lutein, α-carotene and β-carotene than women in the other quintiles. The lower carotenoid levels associated with higher body weight, despite absence of difference in intake, suggest either that the relationship between intake and serum values are not linear or that intake values were overreported. As the correlation between plasma levels was somewhat stronger with BMI index than with underreporting index, the lower serum levels in the heavier 20% of the population is probably physiological. The non-linear relationship

between intake and serum levels of carotenoids may be related to different turnover rates or to different body fat content of overweight individuals.[38–41]

Despite often expressed doubts about the accuracy of the weighed intake technique, it was remarkably well correlated with two biological markers of food intake, the 24-hour urine nitrogen and potassium (Table 5). This was true even when results were considered separately for those who gave valid results and those who did not, as assessed by the urine to dietary N ratio. To examine possible time-related effects in the protocol, results from all methods and from weighed records obtained at the end of the study were compared with urinary N excretion from the first 24-hour urine collections obtained at the start of the study. Urinary N excretion was also compared with dietary N in a subset of women who had completed all four dietary methods. In all cases, the same pattern was seen, in that results from the weighed records gave highest correlations with biological variables, and those from the FFQ and 24-hour recall the lowest. The next best method was the 7-day estimated food diary or record. Energy adjustment improved the agreement between results from the FFQ and 24-hour urine nitrogen and potassium, but did not affect the overall ranking of methods in their ability to estimate habitual diet as assessed from biomarkers. Whilst it may be thought that results from this study, with volunteers from a selected population sample, may not be applicable to other populations, or less highly motivated individuals, studies elsewhere in the UK, USA, Scandinavia and Italy have also shown that records of food consumption are more likely than other methods such as FFQ to rank subjects more accurately when estimating habitual diet from biomarkers.[33,35,42,43]

Furthermore, despite the selected nature of participants in the validation study, the results from a random sample of all participants in the UK EPIC cohort are broadly similar. The FFQ was improved by asking, for example, separate questions about milk and breakfast cereal consumption, and Table 7 shows that some mean differences in nutrient results obtained from the improved version of the FFQ and the diary were reduced. With only a single blood sample, biomarker comparisons were somewhat limited, but nevertheless the same overall pattern as was found in the initial validation study were seen in this initial set of results from the EPIC cohort itself. Correlations between serum carotenoids and estimates of carotenoid intake were rather higher when using dietary intake data from the diary than those obtained with the FFQ. More detailed biomarker information is presently being collected for further validation studies.

The errors associated with known fluctuations in the frequency of consumption of micronutrients such as vitamin C and the carotenes are reduced by repeated observations with 24-hour recall or record methods, but the FFQ is designed to assess usual intake without the need for repeated assessments. However, correlations between plasma levels and estimates of intake of vitamin C intake were as high when derived from a single 24-hour recall as those obtained from the FFQ and the 7-day diary (Tables 6, 7). Correlations with plasma values for both vitamin C and carotenoids were highest when intake values from weighed records were used (Table 6). Only the correlations between α-carotene and dietary carotene from the FFQ approached those found with weighed records. In carrots α carotene represents a greater proportion of the total carotene than in other foods and the reason for the best correlation may have related to the ease with which individuals are able to quantify carrot consumption. The correlations were less good with the major source in food, β-carotene ($r = 0.14$) which is derived from several foods. This finding of higher correlations between dietary carotene and plasma α-carotene than plasma β-carotene has been shown before.[25,33] Direct comparisons with the estimated menu record (7-day diary) were not possible, because plasma carotenoids were not analysed in this group. However, data from a similar method, a 7-day checklist[5] were available. In general, the correlations between carotenoids estimated from the 7-day checklist records were higher than those obtained from the FFQ, for example 0.15 for β-carotene assessed by FFQ, and 0.28 for β-carotene assessed from the record (Table 6).

Overall, the unstructured 7-day diary (estimated record) had the next highest correlation coefficients when compared with biomarkers and was able to classify a greater proportion of individual values into the correct quartile of the distribution when compared with weighed records. No biases in mean intakes of either foods or nutrients were found, and comparisons between results obtained from it and urinary biological markers of intake were almost as good as those obtained from 16 days of weighed records. For these reasons, and because of the flexibility of a food diary in investigating any further hypotheses, the food diary was included as one of the three methods used in the UK EPIC cohort. 93% of diaries are returned, enabling it to be used as the main method for nested case-control studies in the UK arm of the EPIC study, with repeat investigations as the cohort progresses over time.

ACKNOWLEDGEMENTS

The participants, study nurses, co-ordinating centre of EPIC, and participating general practitioners of Norfolk

and Cambridge are gratefully thanked for their assistance. Undergraduate students from Coleraine, Surrey, and Wageningen Universities and Leeds Polytechnic were extensively involved in the collection and interpretation of the field work. Support for this study was obtained from the Medical Research Council, the Cancer Research Campaign, British Heart Foundation, Europe Against Cancer Programme of Commission of European Communities, the Ministry of Agriculture, Fisheries and Food, the Imperial Cancer Research Fund, and the Vegetarian Society.

REFERENCES

[1] Bingham S. The dietary assessment of individuals; Methods, accuracy, new techniques and recommendations. *Nutrition Abstracts and Reviews* 1987; **57:** 705–42.

[2] Callmer E, Riboli E, Saracci R, Akesson B, Lindegarde F. Dietary assessment methods evaluated in the Malmo study. *J Intern Med* 1987; **233:** 53–57.

[3] Bingham S, Cummings J H. The use of 4 amino benzoic acid as a marker to validate the completeness of 24 h urine collections in man. *Clin Sci* 1983; **64:** 629–35.

[4] Bingham S, Cummings J H. Urine nitrogen as an independent validatory measure of dietary intake. *Am J Clin Nutr* 1983; **42:** 1276–89.

[5] Bingham S A, Gill C, Welch A *et al*. Comparison of dietary assessment methods in nutritional epidemiology *Br J Nutr* 1994; **72:** 619–43.

[6] Bingham S A, Cassidy A, Cole T *et al*. Validation of weighed records and other methods of dietary assessment using the 24 h urine technique and other biological markers. *Br J Nutr* 1995; **73:** 531–50.

[7] Paul A A, Southgate D A T. McCance and Widdowson's *The Composition of Foods*. London: Her Majesty's Stationery Office, 1978.

[8] Wiles S J, Nettleton P A, Black A E, Paul A A. The nutrient composition of some cooked dishes eaten in Britain: a supplementary composition table. *J Hum Nutr* 1980; **34:** 189–223.

[9] Bingham S A. Non starch polysaccharide intake of a representative sample of British Adults. *J Hum Nutr Dietetics* 1990; **3:** 333–37.

[10] Bingham S, Williams D R R, Cole T J, Price C P, Cummings J H. Reference values for analytes of 24 h urines known to be complete. *Ann Clin Biochem* 1988; **25:** 610–19.

[11] Schofield W N, Schofield C, James W P T. Basal Metabolic Rate. *Human Nut: Clinical Nut* 1985; **39c (Suppl. 1):** 1–96.

[12] Margolis S A, Davis T P. Stabilization of ascorbic acid in human plasma, and its liquid chromatographic measurement. *Clin Chem* 1988; **34:** 2217–23.

[13] Bates C, Cowen T D. Effects of age and dietary vitamin C on the contents of ascorbic acid in lens and aqueous humour of guinea pigs. *Exp Eye Res* 1988; **46:** 937–45.

[14] Thurnham D I, Smith E, Flora P S. Concurrent liquid chromatographic assays of retinol, a tocopherol, b carotene, a carotene, lycopene and cryptoxanthin in plasma with tocopherol acetate as internal standard. *Clin Chem* 1988; **34:** 377–81.

[15] Willett W C, Sampson L, Stampfer M. Reproducibility and validity of semiquantitative food frequency questionnaire. *Am J Epidemiol* 1985; **122:** 51–65.

[16] Willett W C, Sampson L, Brown M L *et al*. The use of a self administered questionnaire to assess diet four years in the past. *Am J Epidemiol* 1988; **127:** 188–99.

[17] National Food Survey Committee (1982). *Annual Report of Household Food Consumption and Expenditure* 1980, London: Her Majesty's Stationery Office.

[18] Crawley H. *Food Portion Sizes*. London: Her Majesty's Stationery Office, 1988.

[19] Braddon F E M, Wadsworth M E J, Davies J M C, Cripps H A. Social and regional differences in food and alcohol consumption in Britain. *J Epidemiol Community Health* 1988; **42:** 341–49.

[20] Key F B, Paul A A, Harter A C, Price G M, Cole T J, Wadsworth M E J. A prospective method for dietary assessment of a population sample using DIDO, a direct computer data entry system. (Unpublished).

[21] Edington J, Thorogood M, Geekie M, Ball M, Mann J. Assessment of nutritional intake using dietary records with estimated weights. *Hum Nutr Dietetics* 1989; **2:** 407–14.

[22] Holland B, Unwin I D, Buss D H. Cereals and Cereal Products, Third Supplement to McCance and Widdowson's Composition of Foods (4th edn). London: Her Majesty's Stationery Office, 1988.

[23] Holland B, Unwin I D, Buss D H. Milk Products and Eggs, Fourth Supplement to McCance and Widdowson's Composition of Foods (4th edn). London: Her Majesty's Stationery Office, 1989.

[24] Edmond L M, Thurnham D, Oakes S, Bingham S. The stability of plasma vitamin C during storage in cryo-bio straws in liquid nitrogen and in MPA. (Unpublished).

[25] Gregory J, Foster K, Tyler H, Wiseman M. The Dietary and Nutritional Survey of British Adults. London: Her Majesty's Stationery Office, 1990.

[26] Bingham S A, Plummer M, Day N E. Energy adjusted intakes. *Br J Nutr* 1995; **74:** 141–43.

[27] Bingham S, Day N E. Use of biomarkers to validate dietary assessments and the effect of energy adjustment. *Am J Clin Nutr* (In Press), 1996.

[28] Willett W C. *Nutritional Epidemiology*. New York: Oxford University Press, 1990.

[29] Block G. A Review of Validations of Dietary Assessment Methods. *Am J Epidemiol* 1982; **115:** 492–505.

[30] Margetts B M, Nelson M (eds). *Design Concepts in Nutritional Epidemiology*. New York: Oxford University Press, 1991.

[31] Goldbohm R A, van den Brandt P A, Brants H A M, van't Veer P, Strumans F, Hermus R J J. Validation of a dietary questionnaire used in a large scale prospective cohort study on diet and cancer. *Eur J Clin Nutr* 1994; **48:** 253–65.

[32] Rimm E B, Giovannucci E L, Stampfer M J, Colditz G A, Litin L B, Willett W C. Reproducibility and validity of an expanded self administered semiquantitative food frequency questionnaire among male health professionals. *Am J Epidemiol* 1992; **135:** 1127–32.

[33] Yong L C, Forman M, Beecher G R *et al*. Relationship between dietary intake and plasma concentration of carotenoids in premenopausal women. *Am J Clin Nutr* 1994; **60:** 223–30.

[34] Delcourt C, Cubeau J, Balkau B, Papoz L. Limitations of the correlation coefficient in the validation of diet assessment methods. *Epidemiology* 1994; **5:** 518–24.

[35] Rothenberg, E. Validation of the FFQ with the 4 day record method and analysis of 24 h urinary nitrogen. *Eur J Clin Nutr* 1994; **48:** 725–35.

[36] Krebs-Smith S M, Heimendinger J, Subar A F, Patterson B H, Pivonka E. Estimation of fruit and vegetable intake using FFQ: a comparison of instruments (Abstract). *Am J Clin Nutr* 1993; **59 (Suppl. 1):** 283S.

[37] Goldberg G R, Black A E, Jebb S A *et al*. Critical evaluation of energy intake data: 1 Derivation of cut off limits to identify underrecording. *Eur J Clin Nutr* 1991; **45:** 569–81.

[38] Connett J E, Kuller L H, Kjelsberg *et al*. Relationships between carotenoids and cancer. *Cancer* 1989; **64:** 126–34.

[39] Henderson C T, Mobarhan S, Bowen P. Normal serum response to oral beta carotene in humans. *J Am College Nutr* 1989; **8:** 635–35.

[40] Rock C L, Sweindseid C. Plasma carotenoids in anorexia nervosa and obese patients. *Methods in Enzymology* 1993; **214:** 116–23.

[41] Willett W C, Polk F, Underwood B *et al*. Relation of serum vitamins A and E and carotenoids to the risk of cancer. *New Engl J Med* 1984; **310:** 430–34.

[42] O'Donnell M G, Nelson M, Wise P, Walker D. A computerised diet questionnaire for use in diet health education. *Br J Nutr* 1991; **66:** 3–15.

[43] Porrini M, Gentile M, Fidanza, F. Biochemical validation of a self administered semi quantitative FFQ. *Br J Nutr* 1995; **74:** 323–33.