

Nutritional methods in the European Prospective Investigation of Cancer in Norfolk

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Abstract

Objective: To describe methods and dietary habits of a large population cohort.

Design: Prospective assessment of diet using diet diaries and food-frequency questionnaires, and biomarkers of diet in 24-h urine collections and blood samples.

Setting: Free living individuals aged 45 to 75 years living in Norfolk, UK.

Subjects: Food and nutrient intake from a food-frequency questionnaire on 23 003 men and women, and from a 7-day diet diary from 2117 men and women. Nitrogen, sodium and potassium excretion was obtained from single 24-h urine samples from 300 individuals in the EPIC cohort. Plasma vitamin C was measured for 20 846 men and women.

Results: The food-frequency questionnaire (FFQ) and the food diary were able to determine differences in foods and nutrients between the sexes and were reliable as judged by repeated administrations of each method. Plasma vitamin C was significantly higher in women than men. There were significant ($P < 0.001$) differences in mean intake of all nutrients measured by the two different methods in women but less so in men. The questionnaire overestimated dairy products and vegetables in both men and women when compared with intakes derived from the diary, but underestimated cereal and meat intake in men. There were some consistent trends with age in food and nutrient intakes assessed by both methods, particularly in men. Correlation coefficients between dietary intake assessed from the diary and excretion of nitrogen and potassium in a single 24-h urine sample ranged from 0.36 to 0.47. Those comparing urine excretion and intake assessed from the FFQ were 0.09 to 0.26. The correlations between plasma vitamin C and dietary intake from the first FFQ, 24-h recall or diary were 0.28, 0.35 and 0.40.

Conclusions: EPIC Norfolk is one of the largest epidemiological studies of nutrition in the UK and the largest on which plasma vitamin C has been obtained. Methods for obtaining food and nutrient intake are described in detail. The results shown here for food and nutrient intakes can be compared with results from other population studies utilising different methods of assessing dietary intake. The utility of different methods used in different settings within the main EPIC cohort is described. The FFQ is to be used particularly in pooled analyses of risk from diet in relation to cancer incidence within the larger European EPIC study, where measurement error is more likely to be overcome by large dietary heterogeneity on an international basis. Findings in the UK, where dietary variation between individuals is smaller and hence the need to use a more accurate individual method greater, will be derived from the 7-day diary information on a nested case–control basis. 24-h recalls can be used in the event that diary information should not be forthcoming from some eventual cases. Combinations of results utilising all dietary methods and biomarkers may also be possible.

Keywords

Diet

Cancer

Prospective

Biomarkers

Food-frequency questionnaires

Urine

Food diaries

Epidemiological estimates suggest that on average 32–35% of cancers could be prevented by changes in diet, although the estimated contribution of diet varies from as low as 10% for lung cancer to 80% for cancer of the breast, prostate and large bowel^{1,2}. Many dietary items have been suggested to promote or prevent cancer, but in two recently published reviews, evidence for only a few specific relationships was found^{3,4}. The aim of the European Prospective Investigation of Cancer and Nutrition (EPIC) is to improve the definition of diet–cancer associations using prospective, large-scale data with improved methods for dietary assessment including biological markers. The interaction between nutritional and genetic factors (and with other environmental factors such as exercise and smoking) will also be studied⁵.

Within EPIC, the scope of the Norfolk arm has widened from cancer to include other diseases of middle- or late-age onset with strong dietary components, such as cardiovascular disease, diabetes, osteoporosis, and possibly dementia. The purpose of this paper is to describe the nutritional methods from the Norfolk arm of EPIC, one of the largest nutritional studies conducted in the UK. To allow a comparison of the different methods used in the study, information on nutrient and food consumption in this large population is shown, together with results from limited studies on the validity of dietary methods incorporating biomarkers of nutritional intake.

Methods

Core protocol of EPIC

EPIC has a core protocol followed in each of the nine different collaborating countries (Sweden, UK, Denmark, Netherlands, Germany, France, Greece, Italy and Spain). Recruitment is now virtually complete, blood having been collected from about 370 000 individuals and questionnaire and nutritional data from about 470 000. All centres carried out initial validation studies of nutritional methods with reference methods and biological markers prior to embarking on recruitment. Results of these validation studies have been reported elsewhere⁶.

Protocol of EPIC Norfolk

The UK EPIC cohort comprises two arms: one managed in Cambridge which has recruited participants from Norfolk and one managed in Oxford which includes UK participants, of whom about 50% are vegetarian. Similar methods for dietary assessment are used in both, but results for the Cambridge procedures are reported here.

Norfolk was chosen by the Cambridge-based group because it is a geographically defined area covered by the East Anglian Cancer Registry housed at the Institute of Public Health in Cambridge. The city of Norwich and surrounding small towns and rural areas were chosen as the study area, and 35 medical practices agreed to participate. With certain exclusions such as terminal

illness, all individuals aged 45 to 74 years registered with these practices in Norfolk were approached by the general practitioners and asked if they would like to participate in the EPIC study. Recruitment began in March 1993 and was completed at the end of 1997. In total 77 630 individuals were invited to participate, of whom some information (including a written consent to the trial and a single 24-h dietary recall, see below) was obtained on 30 452 at the close of recruitment. Ethical permission for the study was obtained from The Norfolk and Norwich Hospital Ethics Committee. Participants gave permission for the general practitioners to provide information from medical records, and to attend for a medical examination at which blood was to be taken that could be used for research at a later date. At the close of recruitment, 25 630 individuals had attended the medical examination, of whom blood samples had been taken and stored in liquid nitrogen from 22 756, 89% of attendees.

Preliminary validation studies of dietary methods in the UK EPIC cohort

Validation measures of the accuracy of dietary intake were assessed using biological markers, previously devised from work conducted in the metabolic suite of the Dunn Nutrition Unit⁷. The accuracy of several possible methods for use in EPIC was investigated by comparison with 16-day weighed records and the biomarkers, 24-h urine N and K, plasma carotenoids and plasma vitamin C. As a result of these studies, it was decided to use three methods – 7-day diaries (repeated at 18 months and 3–4 years), an improved food-frequency questionnaire (FFQ) and a 24-h recall – to assess diet in the planned prospective study in Norfolk^{8,9}.

24-h recall

A simple 24-h diet recall was sent out to all potential participants in EPIC Norfolk together with the letter of invitation, the questionnaire covering health and lifestyle events, and a consent form. The 24-h recall took the form of a printed double A4 sheet of paper, and asked for a record of all food and drink, including snacks, taken over the previous day. A written example was included. 30 452 completed 24-h recalls were returned to the Cambridge centre by post for later coding using an in-house computer program, see below.

Food-frequency questionnaire

Following receipt of the completed 24-h recall and signed consent form, an appointment was made for the participant to be seen by a nurse at the GP surgery or a clinic in central Norwich. Together with information about the appointment, a food-frequency questionnaire was posted to participants for completion prior to the clinic or surgery visit. At the close of recruitment, 25 630 individuals had completed a medical examination and an

FFQ. In-house computer programs have been developed for the data entry and analysis of these questionnaires.

The FFQ comprised a list of 130 foods, similar to that illustrated elsewhere¹⁰. Revisions were however introduced for milk and breakfast cereals. In the revised version, the amount of milk consumed per day was specified to the nearest quarter of a pint, and the brand name and type of most often used breakfast cereals was listed. In addition, a number of supplementary questions were asked in order to better categorise total fat and fatty acid consumption, such as selections for the fat most often used for frying and for baking and amount of fat usually eaten on meat. Answers to these questions were mapped back to the relevant item on the list. Accompanying the list was a multiple response grid in which respondents estimated how frequently foods were eaten on average over the past year. Respondents were asked to choose one of nine categories of response, ranging from never to more than six times per day. Responses were coded 1–9 in data entry. Missing choices or two or more choices per line were omitted from the analysis. FFQs were considered incomplete if the frequency of 10 or more food items was missing and these were excluded from analysis. Portion weights and representative food codes were assigned to each item, with no distinction according to sex or age for portion weights, largely as those used in the initial validation study.

7-day dietary diary

The 7-day food diary was based on that used in the previous validation study but was printed in colour. It comprised an A5, 45-page booklet in which the description, preparation and amount of foods eaten at main meals, snacks and between meal times over seven consecutive days could be recorded¹⁰. Detailed instructions on the information required in describing and quantifying each type of food or drink were printed on the front pages of the booklet. This encouraged participants to include information on cooking method, type of fat or oil used in cooking, brand name of commercial products or recipes used in cooking. The instructions also provided suggestions for describing the amounts eaten, which might be useful for different foods, often using convenient household measures such as tablespoons, bowls, glasses, numbers, units, slices or packet weights. Seventeen sets of colour photographs were included to help the participants describe the portion size of the food they consumed. Each set showed a small, medium and large portion of the different foods.

At the conclusion of the medical examination, see below, the nurse spent approximately 15 minutes explaining the layout of the diary and instructing participants how to fill out the record. To assist in the explanation, the nurse asked the participant to recall the previous day's intake, from waking until going to bed, and this description was written into the first day of the

diary. A carbon copy of this first 24-h recall was retained by the nurse. The subject was advised to check specific food types, brand names and recipes on their return home and to add this to the day's record completed with the nurse where necessary. The importance of fully describing and quantifying all food and drink consumed was emphasised, so that sufficient information was available to accurately code and analyse the food diaries at a later date. At the conclusion of the interview, the participant was asked to continue recording their food intake for the next six days. Participants were given a Freepost envelope for return of the diaries after the recording period. 23 656 diaries, 92% of those issued, have been returned.

A suite of in-house programs and databases, Data Into Nutrients for Nutritional Research (DINER), was developed to convert diary information into nutrient data. The descriptions and portion size estimates used in the diary were converted into weights and this information is linked to codes for tables of food composition. The tables can be updated and extended as new data become available^{11–20}. The system was designed to avoid loss of the detail given in the diary during coding and matching to published food codes and to cope with the rapidly changing UK food supply. Compared with The Royal Society of Chemistry published data on 2460 foods, the DINER system contains data for approximately 7190 additional foods that were identified as necessary to match the detailed information in food diaries. The 'new' or additional foods were mapped to proportions of between one and four published codes to match the known nutritional composition as best as possible until more precise data are available. The nutritional composition of the additional food was derived from available information, manufacturers' data or calculated from recipes. For example, the database currently includes approximately 50 codes for muesli, covering the different supermarket and branded products recorded in the food diaries. These were linked (singly or proportionately, depending on composition) to the three codes for muesli published by The Royal Society of Chemistry to match their individual composition as best as possible. The type of fat used was selected and mapped for certain foods such as fried foods and baked items.

Portion sizes were coded as described in the diary for each item, using a large database containing information on the portion sizes of specific foods. The large number of possible combinations for each food was rationalised by assigning some weights at the group or database level; for example, all margarines and spreads were assigned the same weight when spread on bread. Within the DINER program, each food group had a specific range of portions available, all of which have a quantity attached. The portion weight data were obtained from direct measurements of foods and from published sources^{21,22}. As new portions became necessary they were added to

the system. Descriptions of volume (for example when photographs were used to describe portion sizes of permitted items) were converted to weight in the program by reference to a database containing the density values for many foods derived from direct measurements or from available data. The weight of the food item was adjusted for inedible waste and gains and losses in cooking. A manual for reference for coders was developed to standardise procedures for data entry. All entered and coded data were checked. On average each diary took 2.0 hours to code, including checking and editing of the entered data. In addition, a further 1.0 hours' work was required per diary in creating new food codes and mapping of nutritional data, running check programs and cleaning the coded data, and running the analysis programs followed by further checks on the data.

The medical examination and other information

The medical examination at a clinic or general practitioner surgery lasted for approximately 45 minutes, during which time height (without shoes), weight (light clothing), waist, hip and chest measurements were taken using standardised methods²³. Basal metabolic rate (BMR) was calculated from body weight²⁴.

Blood (42 ml) was withdrawn using a tourniquet and Safety Monovettes (Sarstedt, Numbrecht, Germany). Blood for vitamin C analysis was collected in 10 ml tubes containing 1 ml of 3.13% trisodium citrate-2-hydrate solution, and stored overnight in a refrigerator prior to processing in a laboratory. Plasma (0.25 ml) for vitamin C analysis was preserved in 0.5 ml of 10% metaphosphoric acid at -80°C and analysed within a week at Addenbrookes Hospital Clinical Biochemistry Department in Cambridge by fluorometric assay using a Monarch centrifugal analyser²⁵. Previous studies showed that the overnight storage procedure resulted in a small (7%, $P = 0.012$) reduction in plasma vitamin C and a high correlation (0.84) between results from freshly processed and stored samples²⁶. Results shown here have not been corrected for dilution arising from the use of citrate in the collection tubes.

Validation studies with biomarkers and studies on repeatability

Plasma vitamin C was measured at baseline as described above. Intakes of vitamin C were not corrected for consumption of vitamin C supplements. Single 24-h urine collections were collected from participants' homes three to 18 months after recruitment. The 24-h urinary excretion of nitrogen, potassium and sodium was analysed in these samples. Details of the 24-h urine collection procedure and use of *p*-aminobenzoic acid (PABA) for checking on the completeness of 24-h urine samples is described elsewhere⁸. At 18 months, all of those who had attended the medical examination (except those who had died or withdrawn from the study) and who had completed the food diary were approached by post and asked to complete a second food diary and a questionnaire to ascertain details of changes in health since recruitment. Those who did not complete the food diary were sent a repeat 24-h recall. One reminder was sent to non-responders.

Statistical methods

Means and standard deviations were obtained using SAS version 6.12. Differences in means were assessed by unpaired *t*-tests. Trends in nutrient and food intake by age were determined from regression analysis using STATA version 6.

Results

Samples studied in relation to the main EPIC cohort

Table 1 shows the age structure of the main EPIC cohort. Ninety-three per cent of individuals recruited were in the age range 45–74 years old. Some 23 003 FFQs from individuals in this age range were available for analysis. Table 1 shows that the age structure of this FFQ sample was similar to that of the main cohort. Plasma vitamin C was available from 11 314 women and 9532 men. Table 1 shows that the sample for whom food diary data were available contained rather more participants in the 65–74 age range and fewer in the 45–54 age range compared

Table 1 Characteristics of the samples studied according to age group (years)

	Total	Women					Men				
		35–44	45–54	55–64	65–74	75–80	35–44	45–54	55–64	65–74	75–80
Main cohort											
Numbers	25 530	536	4820	4255	3875	489	391	3641	3586	3477	460
Percentage		2.1	18.9	16.7	15.2	1.9	1.5	14.3	14.0	13.6	1.8
FFQ sample											
Numbers	23 003	–	4717	4163	3700	–	–	3562	3503	3358	–
Percentage			20.5	18.1	16.1			15.5	15.2	14.6	
Diary sample											
Numbers	2117	–	334	423	483	–	–	140	328	409	–
Percentage			15.8	20.0	22.8			6.6	15.5	19.3	
Biomarker sample											
Numbers	300	25	65	41	36	6	17	41	33	30	6
Percentage		8.3	21.7	13.7	12.0	2.0	5.7	13.7	11.0	10.0	2.0

with the main cohort. Table 1 shows that the subjects from whom a 24-h urine sample had been obtained for biomarker analysis contained a higher percentage of 35–44-year-olds but that otherwise the age distribution was similar to the main cohort.

Nutrient intake, anthropometric and vitamin C measurements assessed by food-frequency questionnaire and by 7-day diary

Table 2 shows dietary intake for selected nutrients as assessed by the food-frequency questionnaire in the 23 003 EPIC participants by sex and by age band. When all ages were combined, reported intakes of nearly all nutrients were significantly greater in men than women, $P < 0.001$, although intakes of non-starch polysaccharides (NSP) and vitamin C were significantly less ($P < 0.001$). Plasma vitamin C levels were also higher in women than men ($P < 0.001$). There was little (but significant) difference in body mass index (BMI) between men and women. Despite their higher reported energy intake, the ratio of energy intake to BMR was significantly lower in men than in women ($P < 0.001$).

Regression coefficients (not shown) of intake against age were significant ($P < 0.001$) for most nutrients, with intakes generally increasing with age (apart from protein, calcium and potassium, which showed no trends) in women. In men, reported intakes of energy, protein, fat, calcium and potassium decreased significantly with age, and intakes of sugars, NSP, vitamin C, vitamin A and carotene increased. In both men and women, reported intakes of alcohol decreased with age. The energy intake to BMR ratio increased with age in both sexes; $P < 0.001$.

Table 3 shows nutrient intakes from the 2117 diaries. When all ages were combined, reported intakes of nearly all nutrients were significantly greater in men than women, $P < 0.001$, except for vitamin C, which was significantly less ($P < 0.001$). As before, plasma vitamin C levels were lower in men than women ($P < 0.001$). There was no significant difference in BMI between men and women. The energy intake to BMR ratio was higher in men than women ($P < 0.001$).

Regression coefficients (not shown) of intake against age were showed a significant ($P < 0.038$) decline with age for most nutrients in both sexes except for vitamin A, which increased with age in women. There were no significant trends with age in sugars and carotene in women, and carotene, vitamin A and vitamin C in men. Plasma vitamin C decreased, as did the energy intake to BMR ratio; BMI increased with age.

When all age-reported intake data from the FFQs from Table 2 were compared with that from the food diary in Table 3, reported intakes of all nutrients were significantly ($P < 0.001$) greater in women. In men, reported intakes in energy, fat, carbohydrate, vitamin A and energy intake to BMR ratio reported by the FFQ were not significantly different from those reported by the diary. Intakes of all

other nutrients were significantly ($P < 0.001$) greater. To take account of age differences, paired *t*-tests were conducted on logged data from the 2062 individuals for whom both diary and FFQ data were available. All differences in nutrient intakes were significant ($P < 0.001$) except for energy ($P > 0.402$) and carbohydrate ($P > 0.132$) in men.

Food intake assessed by food-frequency questionnaire and by 7-day diary

Table 4 shows food intakes as assessed by the food-frequency questionnaire. Reported intakes of most food groups were significantly greater in men than women, $P < 0.001$, apart from no difference in breakfast cereals, fish, nuts, offal, and sauces, and greater ($P < 0.001$) reported intakes of cheese, fruit and other drinks by the women. Regression coefficients (not shown) of intake against age showed significant ($P < 0.016$) increases with age for most food groups in women (except other cereals, meat and alcohol, which declined significantly with age). In men, trends in food group intakes were similar to those in women, apart from bread, fish and milk, which did not change.

Table 5 shows intakes of foods assessed by the 7-day diary in g day^{-1} by age and sex. When all ages were combined, reported intakes of most food groups were significantly greater in men than women, $P < 0.001$, apart from no difference in vegetables and offal, and greater ($P < 0.001$) reported intakes of other dairy products and fruit by the women. In contrast to the FFQ, trends with age were generally not significant, except for intakes of bread, other cereals, meat products and alcohol, which declined significantly ($P < 0.01$) with age. Reported intakes of breakfast cereal and margarine, butter and spreadable fats increased significantly with age in women.

When average reported intakes of food groups from the FFQ (Table 4) were compared with that from the food diary (Table 5), estimates of average food intake by the two methods were similar for some foods, such as bread, breakfast cereals, other cereals, meat, meat products, offal, fish, potatoes, nuts, tea, alcoholic drinks, soups, sauces and miscellaneous foods. However, the questionnaire markedly overestimated ($P < 0.001$) the intake of milk, cheese, eggs, margarine, butter and spreadable fats, vegetables and fruit in women. In men, the questionnaire also overestimated milk, cheese, eggs, margarine, butter and spreadable fats, vegetable and fruit intake, whereas bread, cake, breakfast cereal, other cereal and meat intakes were underestimated ($P < 0.001$) by the questionnaire compared with the diary. Estimates of alcoholic drink and soup intake were similar ($P > 0.05$). To take account of age differences, paired *t*-tests were conducted on logged data from the 2062 individuals for whom both diary and FFQ data were available. All differences in food group consumption were significant ($P < 0.001$) except for alcohol, no difference between both sexes

Table 2 Daily intakes of nutrients by age and sex assessed by food-frequency questionnaire in the Norfolk cohort, energy intake to BMR ratio, body mass index and plasma vitamin C: means and standard deviations

Age (years)	Women		Men		<i>P</i> Men vs. Women	Women						Men					
	45–74		45–74			45–54	55–64	65–74	45–54	55–64	65–74						
Numbers	12 580		10 423			4714	4163	3700	3562	3503	3358						
Energy (MJ)	8.08	2.29	9.17	2.64	<0.001	7.99	2.27	8.01	2.29	8.27	2.31	9.31	2.7	9.11	2.62	9.09	2.57
Fat (g)	70.8	27.2	82.7	31.3	<0.001	70.2	27.0	69.3	27.1	73.1	27.4	84.9	32.4	82.0	31.3	81.1	30.0
Protein (g)	81.2	20.9	84.9	21.9	<0.001	81.1	20.9	81.1	21.1	81.5	20.7	85.6	22.5	84.8	21.6	84.4	21.5
Carbohydrate (g)	244	76	269	87.3	<0.001	239	75	244	76	251	77	269	88	268	87	271	87
Starch (g)	110	39	126	45	<0.001	110	39	109	38	111	39	129	46	125	44	125	43
Sugars (g)	128	48	136	53	<0.001	123	47	129	48	133	49	133	52	137	54	140	55
Alcohol (g)	5.5	8.4	12.2	16.1	<0.001	6.2	8.9	5.1	8.0	4.9	7.9	14.0	17.2	11.8	16.0	10.8	14.8
NSP (g)	18.8	6.7	18.1	6.4	<0.001	18.3	6.6	19.0	6.6	19.3	6.8	17.6	6.3	18.1	6.4	18.5	6.3
% Energy fat	32	6	33	6	<0.001	32	6	32	6	32	6	33	6	33	6	33	6
% Energy protein	17	3	16	3	<0.001	17	3	18	3	17	3	16	3	16	3	16	3
% Energy starch	22	6	22	5	ns	22	4	22	5	22	5	22	5	22	5	22	5
% Energy sugars	25	6	24	6	<0.001	25	6	26	6	26	6	23	5	24	6	24	6
% Energy alcohol	2	3	4	5	<0.001	2	3	2	3	2	3	5	6	4	5	4	5
Vitamin C (mg)	134	64	112	52	<0.001	130	62	136	63	138	66	106	51	113	52	117	53
Vitamin A (µg)	703	700	783	786	<0.001	678	703	688	692	751	702	743	751	800	838	807	764
Total folate (µg)	328	100	328	96	ns	318	99	329	99	339	104	320	97	329	97	334	94
Carotene (µg)	3104	1753	2800	1450	<0.001	3000	1634	3129	1695	3206	1944	2651	1443	2851	1417	2901	1478
Calcium (mg)	990	289	1037	301	<0.001	989	299	987	285	994	279	1046	314	1039	302	1027	285
Iron (mg)	12.0	4.1	12.5	3.9	<0.001	12.0	4.2	12.0	4.1	12.1	4.0	12.5	4.0	12.4	3.8	12.6	4.0
Potassium (mmol)	99	24	99	23	ns	99	24	99	24	98	28	101	24	99	23	98	22
EI/BMR ratio	1.43	0.42	1.28	0.39	<0.001	1.37	0.40	1.41	0.42	1.52	0.44	1.24	0.37	1.26	0.38	1.33	0.40
BMI (kg m ⁻²)	26.3	4.3	26.6	3.3	<0.001	25.3	4.3	26.6	4.4	26.7	4.2	26.3	3.3	26.7	3.3	26.7	3.2
Plasma vitamin C (µmol l ⁻¹)	58.6	19.9	47.0	18.8	<0.001	59.1	19.1	59.8	20.1	56.6	20.7	48.2	18.6	47.4	18.2	45.4	19.5

Table 3 Daily intakes of nutrients by age and sex assessed by 7-day diary in the Norfolk cohort, energy intake to BMR ratio, body mass index and plasma vitamin C: means and standard deviations

Age (years)	Women		Men		<i>P</i> Men vs. Women	Women			Men								
	45–74		45–74			45–54	55–64	65–74	45–54	55–64	65–74						
Numbers	1240		877			334	423	483	140	328	409						
Energy (MJ)	6.95	1.49	9.06	2.06	<0.001	7.31	1.58	6.89	1.47	6.74	1.40	9.91	2.27	9.29	2.03	8.59	1.88
Fat (g)	62.7	19.2	81.9	29.9	<0.001	65.0	20.6	62.1	18.7	61.7	18.6	90.5	28.2	83.4	23.8	77.8	22.8
Protein (g)	63.9	13.4	79.3	16.8	<0.001	66.5	14.1	64.5	13.1	61.6	12.8	85.3	17.3	80.9	17.6	76.0	15.1
Carbohydrate (g)	208	49	265	70	<0.001	216	51	217	50	202	45	290	75	274	70.7	250	65
Starch (g)	107	27	143	40	<0.001	114	29	107	26	103	26	161	46	148	40	132	34
Sugars (g)	97	33	119	44	<0.001	99	35	96	34	95	30	126	46	121	43	114	43
Alcohol (g)	7.2	10.7	14.8	18.7	<0.001	10.1	13.2	6.2	9.0	6.0	9.9	15.9	18.3	15.4	18.8	14.0	18.7
NSP (g)	13.6	4.3	15.4	5.4	<0.001	14.1	4.5	13.6	4.4	13.1	4.1	16	5	15.7	5.5	14.8	5.2
% Energy fat	33	6	33	5	ns	33	6	33	5	33	6	33	5	33	5	33	5
% Energy protein	16	3	15	2	<0.001	16	3	16	3	16	3	15	2	15	2	15	2
% Energy starch	25	5	25	5	ns	25	5	25	5	25	5	26	5	26	4	25	5
% Energy sugars	22	6	21	6	<0.001	22	6	22	6	23	5	20	5	21	6	21	6
% Energy alcohol	3	4	5	6	<0.001	4	5	3	4	3	4	4	5	5	6	5	6
Vitamin C (mg)	87	48	81	47	<0.01	94	55	84	44	84	46	79	44	82	50	82	45
Vitamin A (µg)	594	1096	774	1748	<0.01	401	541	590	1089	730	1342	875	3161	643	1104	845	1468
Total folate (µg)	241	68	291	86	<0.001	250	71	240	66	238	66	300	85	295	94	285	80
Carotene (µg)	1909	1265	2015	1204	ns	2010	1529	1910	1112	1383	1186	2111	1305	1985	1169	2007	1199
Calcium (mg)	765	250	902	274	<0.001	798	260	760	260	748	232	973	304	925	273	858	256
Iron (mg)	10.6	3.7	13.0	4.4	<0.001	10.9	3.6	10.6	3.8	10.3	3.7	13.7	4.7	13.3	4.7	12.5	4.0
Potassium (mmol)	75	17	85	18	<0.001	87	17	75	16	72	16	92	19	87	18	82	17
EI/BMR ratio	1.23	0.27	1.28	0.29	<0.001	1.25	0.28	1.21	0.27	1.24	0.27	1.31	0.31	1.29	0.29	1.25	0.28
BMI (kg m ⁻²)	26.4	4.2	26.5	3.1	ns	25.7	4.0	26.6	4.3	26.6	4.3	26.5	3.3	26.3	3.0	26.6	3.2
Plasma vitamin C (µmol l ⁻¹)	58.5	20.6	47.4	18.6	<0.001	60.5	19.1	58.8	20.7	56.7	21.3	49.1	17.9	49.9	17.5	44.5	19.3

Table 4 Daily intakes of food groups (g day⁻¹) by age and sex assessed by food-frequency questionnaire in the Norfolk cohort: means and standard deviations

Age (years)	Women		Men		<i>P</i> Men vs. Women	Women						Men					
	45-74	45-74	45-74	45-74		45-54	55-64	65-74	45-54	55-64	65-74	45-54	55-64	65-74			
Numbers	12 580	10 423				4714	4163	3700				3562	3503	3358			
Bread	74	52	90	60	<0.001	71	52	74	51	78	53	90	63	80	60	90	57
Cakes	23	28	31	38	<0.001	19	24	22	28	28	32	29	38	30	38	34	38
Breakfast cereals	38	46	38	53	ns	34	42	39	46	43	51	31	43	39	58	45	56
Other cereals	81	55	84	57	<0.001	90	56	78	54	72	51	95	61	83	55	76	53
Milk	338	160	368	173	<0.001	333	164	340	161	343	155	367	180	369	174	368	163
Cheese	33	28	25	22	<0.001	34	30	33	28	30	27	26	22	26	23	24	20
Other dairy	51	46	41	42	<0.001	50	47	52	46	51	46	39	41	42	44	42	41
Eggs	25	19	27	22	<0.001	26	19	25	19	25	19	27	23	27	22	26	21
Spreadable fats	22	17	27	19	<0.001	21	16	22	17	24	17	26	19	27	20	27	18
Meat	69	39	71	40	<0.001	71	38	69	40	67	38	73	40	70	39	69	40
Meat products	23	19	33	27	<0.001	23	19	23	18	24	19	36	30	33	27	31	22
Offal	2	3	2	4	ns	1.7	3	1.7	3	1.8	3	1.8	3	2	3.8	2	3
Fish	38	26	37	26	ns	38	26	38	25	39	26	37	26	37	26	38	25
Potatoes	115	63	128	71	<0.001	111	58	115	64	120	68	126	71	127	39	131	73
Legumes	58	38	62	38	<0.001	59	37	57	36	58	39	62	36	62	39	61	37
Vegetable dishes	7	13	6	10	<0.001	9	13	7	12	6	13	7	11	6	10	4	9
Other vegetables	220	117	189	102	<0.001	213	58	223	114	224	128	176	100	194	102	198	102
Fruit	277	200	212	165	<0.001	264	203	288	202	282	193	199	158	216	177	221	156
Nuts	3	8	3	9	ns	3	7	3	9	3	10	3	9	3	8	3	10
Sugars	45	41	59	48	<0.001	42	39	45	40	50	45	48	55	59	48	51	62
Tea	1034	383	1068	384	<0.001	1045	402	1041	377	1013	362	1094	411	1065	377	1043	357
Other drinks	132	167	111	145	<0.001	145	179	132	170	113	144	127	156	106	141	98	136
Alcoholic drinks	63	107	202	314	<0.001	75	124	58	99	53	91	247	353	193	303	163	271
Soups	32	46	34	46	<0.001	25	38	33	44	40	55	25	37	34	45	42	55
Sauces	13	14	13	15	ns	15	13	13	15	12	13	15	15	13	16	12	13
Miscellaneous	1	3	1	2	ns	1	3	1	3	1	3	1	2	1	3	1	2

Table 5 Daily intakes of food groups (g day⁻¹) by age and sex assessed by 7-day diary in the Norfolk cohort: means and standard deviations

Age (years)	Women		Men		<i>P</i> Men vs. Women	Women						Men					
	45-74	45-74	45-74	45-74		45-54	55-64	65-74	45-54	55-64	65-74	45-54	55-64	65-74			
Numbers	1240	877				334	423	483				140	328	409			
Bread	76	35	108	51	<0.001	79	37	76	34	73	34	126	59	112	53	100	44
Cakes	36	30	54	48	<0.001	32	28	36	32	39	30	54	52	56	51	52	42
Breakfast cereals	37	46	47	54	<0.001	30	33	39	47	40	53	41	56	48	54	48	54
Other cereals	78	55	99	66	<0.001	91	64	75	51	74	50	118	73	104	67	89	59
Milk	189	74	206	145	<0.01	189	142	187	150	192	132	219	164	206	138	202	142
Cheese	15	15	18	17	<0.001	19	20	13	12	14	13	20	20	19	17	17	15
Other dairy	60	75	45	60	<0.001	70	97	63	67	50	60	43	57	49	59	43	61
Eggs	13	14	18	21	<0.001	14	16	13	14	13	14	17	21	18	21	19	21
Spreadable fats	13	13	18	12	<0.001	12	9	15	9	14	10	19	12	18	11	17	11
Meat	72	49	89	60	<0.001	71	55	77	50	69	45	94	77	89	60	87	52
Meat products	22	19	33	29	<0.001	23	22	23	19	20	16	40	33	34	32	31	24
Offal	1	4	1	4	ns	0.3	2	0.9	4	1.2	4.1	0.8	4.0	0.7	3.3	1	5
Fish	35	36	42	37	<0.001	35	48	36	33	35	29	37	34	42	40	43	35
Potatoes	106	56	139	65	<0.001	109	60	105	53	104	54	148	65	139	67	136	63
Legumes	24	24	31	31	<0.001	25	24	24	22	23	22	41	40	33	31	27	25
Vegetable dishes	20	31	18	28	ns	27	36	21	31	15	27	21	25	19	31	15	27
Other vegetables	97	71	96	67	ns	99	83	97	65	96	66	91	54	95	66	100	71
Fruit	172	128	138	122	<0.001	176	145	166	122	174	121	134	128	137	118	141	124
Nuts	2	5	3	8	<0.01	2	6	2	5	1	4	3	9	4	11	2	6
Sugars	33	30	49	39	<0.001	31	29	33	30	34	28	48	39	47	35	50	42
Tea	1101	465	1162	491	<0.01	1188	529	1078	438	1061	432	1280	622	1132	490	1144	433
Other drinks	307	319	45	59	<0.001	337	323	309	343	283	291	298	328	262	345	227	258
Alcoholic drinks	73	112	252	358	<0.001	109	144	66	99	55	90	309	364	283	417	207	294
Soups	28	51	33	57	<0.05	28	47	26	41	30	60	29	63	28	49	38	60
Sauces	19	26	22	28	<0.02	20	23	21	33	17	22	22	20	22	26	22	31
Miscellaneous	4	13	4	15	ns	5	12	5	16	2	10	7	15	5	15	3	14

Table 6 Pearson correlation coefficients between results obtained for nutrients from methods repeated on two different occasions and obtained with the different methods at the first visit, and between methods and biomarkers in 24-h urine collections and plasma vitamin C

Nutrient	Correlations between repeat methods			Correlations between methods		
	1st vs. 2nd diary	1st vs. 2nd FFQ	1st vs. 2nd 24-h recall	1st diary vs. 1st FFQ	1st diary vs. 1st 24-h recall	1st 24-h recall vs. 1st FFQ
Energy (MJ)	0.78	0.68	0.29	0.50	0.54	0.34
Fat (g)	0.73	0.71	0.27	0.58	0.52	0.39
Nitrogen (g)	0.67	0.52	0.23	0.34	0.46	0.22
Carbohydrate (g)	0.78	0.66	0.35	0.48	0.28	0.28
Sugars (g)	0.75	0.67	0.38	0.53	0.57	0.42
Starch (g)	0.75	0.55	0.45	0.40	0.51	0.23
Alcohol (g)	0.78	0.77	0.60	0.76	0.51	0.51
NSP (g)	0.73	0.66	0.40	0.49	0.60	0.36
Vitamin C (mg)	0.68	0.65	0.50	0.34	0.63	0.35
Retinol (μg)	0.01	0.64	-0.02	0.14	0.07	0.25
Carotene (μg)	0.58	0.65	0.13	0.35	0.33	0.14
Potassium (mmol)	0.72	0.62	0.24	0.44	0.54	0.32
Iron (mg)	0.60	0.66	0.27	0.49	0.55	0.34
Calcium (mg)	0.65	0.67	0.21	0.48	0.43	0.38
	1st diary	2nd diary	1st FFQ	2nd FFQ	1st 24 h-recall	2nd 24-h recall
Diet N and 24-h urine N	0.47	0.42	0.15	0.12	0.37	0.21
Diet K and 24-h urine K	0.36	0.40	0.26	0.24	0.31	0.37
Diet vitamin C and plasma vitamin C	0.40	0.37	0.28	0.42	0.35	0.30

($P > 0.841$), and other cereals in women ($P > 0.075$) and meat ($P > 0.038$) and potatoes ($P > 0.333$) in men.

Repeatability of methods and comparison with biomarkers in blood and urine

Of the sample of 300 for whom a food diary, food-frequency questionnaire, 24-h recall and 24-h urine collection were available for analysis, repeat diaries were obtained from 237, repeat FFQs from 171, and repeat 24-h recalls from 170. There were no significant differences in mean intake between the mean values obtained from the first and second diary, FFQ and 24-h recall assessments (data not shown).

Table 6 summarises Pearson correlation coefficients between results obtained from the first and second application of each method. The 7-day diary was generally more repeatable than the other methods, correlation coefficients between the first and repeat diary ranging from 0.60 for iron to 0.78 for carbohydrate, alcohol and energy. Correlations between results from the first and second FFQ were lower in general; for example, 0.68 for energy and 0.66 for carbohydrate. Results for carotene and retinol were, however, more repeatable using the FFQ than the diary and 24-h recall. The reproducibility of the values for nutrients obtained from the two 24-h recalls was lower than for either the FFQ or the diary.

Table 6 also shows the relation between the different methods at the first application. Correlations were generally substantially lower (0.2 to 0.5) between methods than correlations within methods, with the exception of the correlation for total alcohol consumption assessed by the diary and FFQ (0.76). Correlations

between nitrogen, vitamin C and retinol assessed by the FFQ and the diary and 24-h recall were low.

Two-hundred-and-eighteen 24-h urine samples contained 85–100% PABA and were designated complete. Mean (standard deviation, SD) 24-h outputs in complete collections were 11.0 (2.7) g N, 76 (22) mmol K, 139 (51) mmol Na, 2.0 (0.8) l volume, and 93 (6) % PABA. In the incomplete collections, 24-h outputs were significantly ($P < 0.001$) lower with mean (SD) values of 9.5 (3.1) g N, 62 (24) mmol K, 128 (56) mmol Na, 1.6 (0.6) l volume, and 70 (11) % PABA recovery. Only complete 24-h urine samples were used in the subsequent analyses.

Table 6 shows correlations between mean intakes of nitrogen, potassium and vitamin C, as calculated from the different dietary methods, with 24-h urine nitrogen, potassium and plasma vitamin C. Despite the fact that results were available from only single 24-h urine and blood samples, there were significant correlations between dietary intakes and biomarkers.

Correlations between the 7-day diaries and biomarkers were generally higher than those between the other methods and biomarkers. Inclusion of 24-h urine values that contained between 70 and 85% PABA marker, adjusted to 93% recovery of PABA marker, improved the correlations with nitrogen from the first 7-day diary from 0.47 to 0.50, but had no effect on the correlations between dietary and urinary potassium (data not shown). Correlations between plasma levels of vitamin C and the dietary intake from the first diary and FFQ were 0.40 and 0.28. The 24-h recall performed as well as the FFQ when compared with plasma vitamin C.

To examine the relationship between mean intakes of energy, nitrogen and potassium from different dietary

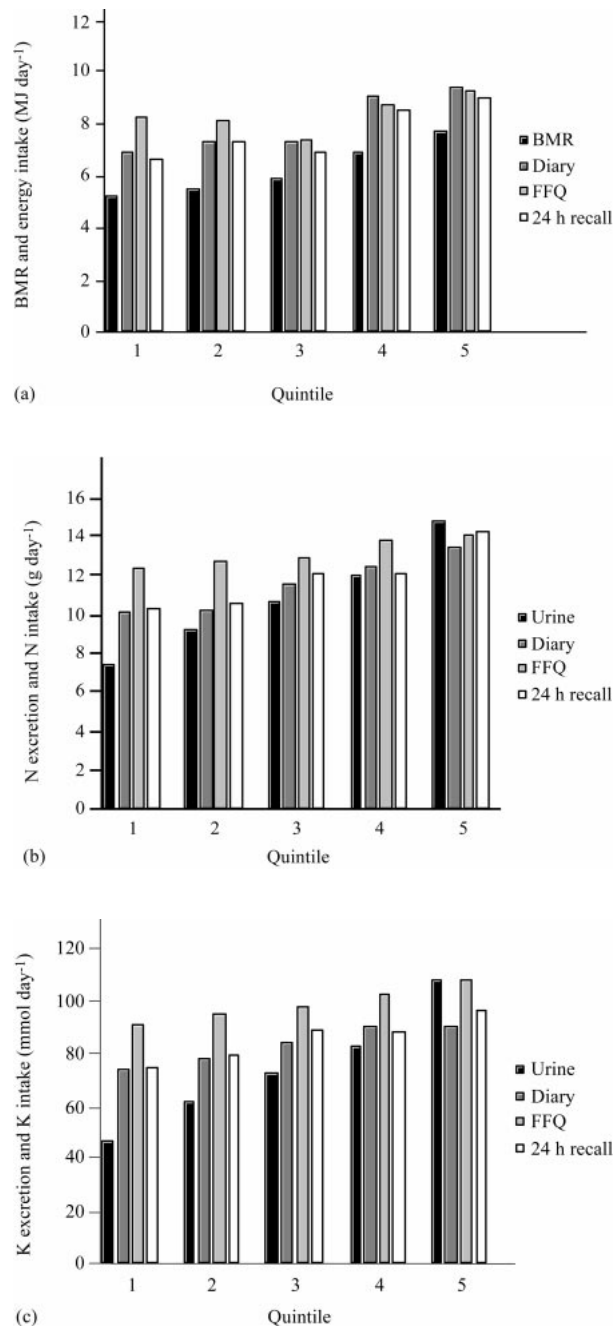


Fig. 1 Comparison of quintiles of: (a) basal metabolic rate and energy intake from different dietary methods, MJ day⁻¹; (b) N excretion from 24-h urine sample and N intake from different dietary methods, g day⁻¹; (c) K excretion from 24-h urine sample and K intake from different dietary methods, mmol day⁻¹. *n* = 300

methods, individuals were arranged into quintiles of basal metabolic rate (in MJ), nitrogen (g) and potassium (mmol) excretion in 24-h urine samples. Figures 1a to 1c show quintiles of BMR, and nitrogen and potassium excretion together with quintiles of intake values from the different dietary methods. Intakes assessed from all three methods increased with increases in BMR and nitrogen and potassium excretion. However the tendency for under-reporting was particularly marked in the fifth quintile and

occurred in all methods. The mean ratio of urine to dietary nitrogen was 0.77 (0.16) in the first quintile, and 1.13 (0.22) in the top quintile using the results from the diary, and 0.66 (0.22) in the first quintile and 1.12 (0.32) in the top quintile using the FFQ. The mean ratio of urine to dietary potassium was 0.68 (0.23) in the first quintile and 1.23 (0.31) in the top quintile using the results from the diary, and 0.56 (0.16) in the first quintile and 1.05 (0.31) in the top quintile using the FFQ. The mean ratio of energy intake to BMR was 1.33 (0.29) in the first quintile and 1.22 (0.23) in the top quintile using the results from the diary, and 1.59 (0.48) in the first quintile and 1.20 (0.39) in the top quintile using the FFQ.

Discussion

The data on food and nutrient intake gathered in EPIC are from one of the largest population surveys of diet currently underway in the UK. It is the largest in which plasma vitamin C has been measured in the UK. When compared with the Health Survey of England, anthropometric, blood pressure and serum lipid variables in the EPIC cohort are similar to those of the national survey, although the cohort has fewer current smokers compared with the general population of England²³. Nevertheless, the cohort is not fully representative of the Norfolk population since only about 40% of those invited to participate did so. Levels of plasma vitamin C in the older groups are also higher than in a national sample²⁸, supporting the view that the EPIC cohort with its low rates for current smoking is healthier than the general population. However, no data on plasma vitamin C are presently available for comparison with a national survey of younger adults²⁹.

Different methods for measuring food intake have been assessed in EPIC Norfolk. Despite very different approaches to measuring food intake, differences between sexes in food and nutrient intake were largely identified by both the FFQ and the diary, and both methods yielded similar intakes of macronutrients when expressed as a percentage of total energy. By both methods, fat intake was comparatively low, at 33% total energy. Fat intakes have been declining in recent years according to household food surveys on a national level³⁰ and in individual studies conducted in the Cambridge area^{8,10,31}, so that this low level when expressed as a proportion of energy intake is probably not attributable to under-reporting. Energy adjustment can lessen differences in reported fat intake between under-reporters and individuals who give valid estimates of nutrient intake³².

In addition, average intakes of foods assessed by the FFQ and by the diary were similar (although statistically different) for most food groups, although in both sexes the FFQ gave significantly higher estimates for vegetables, fruit, margarine, butter and spreadable fats, cheese and milk. Total fruit and vegetable consumption was 515 g as

assessed by the FFQ, for example. This compares with an average intake of 300 g assessed by the food diary. Average consumption of fruits and vegetables by representative samples of free living British adults is even lower, 210 g per day measured by weighed intakes^{28,29}. It is probable that these average intakes of fruit and vegetables assessed by the diary are correct, since, as noted above, plasma vitamin C levels are also higher in the Norfolk cohort compared with a national sample and it is probable that this Norfolk cohort consumes a healthier level of fruits and vegetables than the national average. This tendency of the FFQ to overestimate vegetable, fruit and milk consumption was reported in initial validation studies¹⁰ and has persisted despite changes to the format of the FFQ. Total milk consumption assessed by the FFQ was 350 g per day, but 200 g per day according to the diary. This latter level was similar to that of 230 g found to be consumed in national surveys of free living adults as assessed by weighed intake^{28,29}. Average intake of meat assessed by the diary was 108 g per day compared with 150 g in the national survey of British adults²⁹, and this difference probably reflects a real decline in consumption of these foods over a 10-year period. Meat intake was similar, 110 g day⁻¹, in older free living adults studied recently²⁸.

Table 6 shows that results from methods were correlated to some degree, although the diary gave more repeatable results than the FFQ and 24-h recall. Both methods were able to document consistent changes with age in intakes of other cereals and alcohol (decrease with increasing age) in both sexes, a decrease with age in meat product consumption in men, and an increase with age in spreadable fats and cake consumption in women. The main discrepancies in documentation of age trends between the methods was in bread consumption in women and potato consumption in men, which declined significantly with age according to the diary but increased significantly with age according to the FFQ. Significant increases with age in milk and vegetable consumption in both sexes according to the FFQ were not significant according to the diary. Age trends in nutrient intake were more consistent in men (significant decreases with age for energy, protein, fat, alcohol, calcium and potassium) by both methods, than in women (mostly increases in nutrient intake with age according to the FFQ, but decreasing with age according to the diary). The inability of the FFQ to document declining trends of nutrient intake with age in women suggests that caution is needed when using this method within a single relatively homogeneous cohort. Standard portion sizes were used in the FFQ, and adjustments for age and sex might improve estimates of nutrient intake when using this method.

The energy intake to BMR ratio was low compared with estimates of energy expenditure in both women and men, although somewhat higher when energy intake was assessed by FFQ in the women. All methods therefore

showed a tendency towards under-reporting of energy intake²⁷. This trend was particularly marked in individuals in the upper quintile of basal metabolic rate (Fig. 1a). In women, the diary gave lower estimates of all nutrient intakes compared with the FFQ, whereas in men the diary gave higher estimates of some nutrients compared with the FFQ (Tables 2 and 3).

The validity of methods can be assessed if biomarkers of intake are available such as the 24-h urinary excretion of nitrogen and potassium⁹. For validation of individual assessments, up to 16 days of records and eight complete 24-h urine samples are required⁷. However, only a single collection was made in the present study on participants in the EPIC cohort so that some misclassification into quintiles of potassium and nitrogen excretion would have occurred. Thus the magnitude of the correlation coefficients was lower than in previous studies where repeat 24-h urine samples were collected⁸. Despite this, there was evidence of under-reporting by all methods in the top quintile of N and K excretion (Figs 1b and 1c), and there were evident correlations between individual estimates of nitrogen and potassium and urine excretion of the biomarkers. The previous findings that agreement with urinary biomarkers was better for the 7-day diary than for the FFQ or 24-h recall has been confirmed in this repeat study of EPIC cohort members using only a single 24-h urine collection. The agreement between plasma vitamin C and vitamin C intake was better for the first diary than the FFQ.

EPIC within the UK thus has access to results from three different methods of dietary assessment. The FFQ data are associated with a greater degree of measurement error, as judged by comparison with the independent biomarkers, so that correction factors for regression dilution are substantially greater than those required for the food diary. However, if between-individual variation is increased, correction factors become smaller. Hence the FFQ is to be used particularly in pooled analyses of risk from diet in relation to cancer incidence within the larger European EPIC study, where measurement error is more likely to be overcome by large dietary heterogeneity on an international basis. In addition, findings in the UK, where dietary variation between individuals is smaller and hence the need to use a more accurate individual method is greater, will be derived from the 7-day diary information on a nested case-control basis. All subjects within EPIC Norfolk have dietary information from two 24-h recalls that can be used in the event that diary information should not be forthcoming from some eventual cases. Combinations of results utilising all dietary methods and biomarkers may also be possible.

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