

Joint Health Surveys Unit
March 07

A survey of 24 hour and spot urinary sodium and potassium excretion in a representative sample of the Scottish population

1. Introduction

This study was designed to provide data to establish the progress towards meeting the target to reduce sodium intakes in Scotland, and to compare single spot urine sample collection with 24-hour urine samples with regard to the sodium and potassium concentrations.

Epidemiological, clinical and animal-experimental evidence shows a direct relationship between dietary electrolyte consumption and blood pressure (BP). Furthermore, clinical trials show that a reduction in salt (NaCl) intake reduces BP levels in normotensive and hypertensive populations and prevents the development of hypertension.ⁱ

In the UK, the Committee on Medical Aspects of Food Policy (COMA) panel on Dietary Reference Values (DRV)ⁱⁱ advised that sodium (Na) intakes should be kept below 3.2g (or 8.4g of salt) per day and set the Reference Nutrient Intake (RNI) for men and women at 1.6g of sodium (or 4.2g of salt) per day. Following this, COMA's Cardiovascular Review Group recommended that salt intake should be gradually reduced further to a daily average of 6g.ⁱⁱⁱ This recommendation was accepted in a recent report on salt and health by the Scientific Advisory Committee on Nutrition (SACN) and the Scottish Office's "Eating for health: Diet Action Plan for Scotland" (SDAP) includes the dietary target to reduce the sodium intake of the Scottish population to 100mmol per day (equivalent to 6g of salt). This target for salt is included in the Food Standards Agency Strategic Plan 2005-2010, and the Scottish Executive is committed to implementation of the SDAP well beyond 2010.

The intakes of sodium (Na) and potassium (K) can be estimated by measuring urinary excretion, given that under normal circumstances this is the pathway for their elimination. As electrolyte excretion rates reflect the diet of an individual, unless the diet is very stable over time, variation in Na and K excretion from spot samples taken at different times of day within the same individual can be large, often larger than the variation among a group of individuals in westernised populations. Therefore a single sample is not enough to accurately estimate the true mean intake for an individual, but an earlier study (HSE 2003) has shown that they can be used in epidemiological studies to characterise

the intakes of groups of individuals, provided that they can be correlated with 24-hour measurement.

This study has taken both a 24-hour measurement and three spot samples from each respondent, with the aim of establishing the extent to which findings from the spot samples correlate with those from the 24-hour samples.

2 Methodology

2.1 Recruitment, sample, and response rates

The aim of the study was to achieve 550 usable spot and 24-hour urine samples. The sample of respondents came from two sources. Originally, it was hoped that following up selected respondents to the Scottish Health Survey (SHS) 2003 would yield a large enough sample. This was not the case, so a separate exercise, using random digit dialling, was required to generate and recruit fresh additional sample. In this section both sampling methods are outlined.

Sampling from Scottish Health Survey 2003

Respondents were selected from a sub-sample of those who had taken part in SHS 2003 and who had agreed to be re-contacted. To provide a representative sample of the population of Scotland including the Highlands and Islands, while providing practical and economic work assignments for nurses, SHS sampling points were grouped to provide some clustering and then 34 pairs of points were selected.

A letter was sent to selected respondents from the SHS explaining that the study was taking place and that they would be contacted by telephone. Recruitment for the 24-hour urine study took place by telephone, using NatCen's experienced telephone interviewers who called, explained about the survey and arranged a suitable time for a nurse to visit. Within each household, no more than two adults, aged 19 to 64, were eligible to take part in the study. Response was monitored to ensure that approximately equal numbers of men and women were included. Respondents were offered an incentive of £15 per person on successful completion of the study.

In total, 770 households were sampled, containing 1269 respondents. A relatively high proportion, 25%, of these households could not be contacted (either the telephone number was unusable, or the sampled respondents had moved away with no new address available). At the remaining households, 64% of households, 594 individuals, agreed on the telephone to a nurse visit, and this generated 448 useable samples. This included 6 who provided a 24-hour sample and two out of three spot samples. This represented 34% of all possible individuals in sampled households, which was 102 fewer than anticipated. Therefore, a supplementary sample was required, and for this a telephone sample was generated using random digit dialling.

Table A: Response from sample from the Scottish Health Survey 2003

	Households		Individuals	
	Number	%	Number	%
Total issued	770	100%	1269	100%
Telephone number unusable	107			
Moved, no new address	52			
Total usable	611	75%		
No contact	31			
Refused	174			
Ill, other non-productive	16			
Agreed at telephone interview	390	51%	594	47%
Full productive			441	35%
Partial productive			6	*

Random digit dialling sample

Random digit dialling is a method where a representative sample of telephone numbers is generated at random from a frame of all possible telephone numbers. Many of the numbers generated are non-working numbers (some of which, at the time of the project, could be identified and removed from the sample). Others are non-residential.

NatCen was provided with a RDD sample of telephone numbers from within 17 of the original points covered during the follow up to the SHS 2003. This sample covered all eligible telephone area codes located in these 17 points. The Oftel database lists the first seven digits of all telephone numbers that have been allocated to telephone companies for land lines (e.g. 01222 78XXXX). For each selected area code, the last four digits were randomly generated.

In total, 1,970 telephone numbers were generated (after removing non-working numbers). Telephone interviewers rang these numbers and attempted to recruit up to two adults aged 19-64 to the survey in a similar way to the initial SHS follow up. As there was no existing information on these households or respondents (unlike the SHS respondents, who had taken part in a detailed Health Survey), the telephone interviewers also asked a household questionnaire (which covered basic demographic details and some information on eating habits). Nurses then visited co-operating households.

Table B shows the response analysis for the random digit dialling sample, which generated 192 individuals who agreed to the survey on the telephone, and 126 useable samples. This included 1 who provided a 24-hour sample and two out of three spot samples.

Table B: Response from sample from Random Digit Dialling

	Households		Individuals	
	Number	%	Number	%
Total generated	1970			
Not used	589			
Total issued	1381	100%		
Telephone Number Unusable: technological barriers, fax, modem etc	25			
Out of service, disconnected, non-residential	261			
Unable to confirm if address is residential/eligibility of respondents	413			
Total usable	683	49%		
Not Eligible:				
No-one in HH aged 19-64	299			
Other ineligible	4			
Eligible	379	55%		
Refused nurse appointment	74			
Proxy refusal	182			
Agreed at telephone interview	123	32%	192	100%
Full productive			126	
Partial productive			1	

Overall response

Overall, 574 adults (243 men, 331 women), aged 19 - 64, completed the study and provided spot urine samples and one 24-hour sample (448 from the SHS sample, and 126 from the random digit dialling sample). This included 7 who provided a 24-hour sample and two out of three spot samples. In analysis, 10 samples were discarded because of incomplete or inconsistent labelling, leaving 564 useable samples. Data collection took place between March and November 2006.

3 Nurse training

17 nurses attended a half-day briefing covering the background and purpose of the study, and the methodology. This comprised a teaching and practice session designed to familiarise nurses with the rationale for the study, the methodology and fieldwork procedures. This included a practical demonstration of the Sarstedt syringe that respondents would use for collecting spot samples.

4 Data collection procedures

Ethical approval for the comparison study was granted by the MREC.

The study used the same protocol and procedures that were used in a similar study as part of the HSE 2003. As well as providing urine samples, respondents were asked to take three PABA (para-aminobenzoic acid) tablets at specified intervals. Analysis of PABA excretion provides a measure of the completeness of the 24-hour sample.

The nurse first checked eligibility, and respondents were excluded from the study if they were pregnant; or if they were allergic to hair dye, sunscreen or vitamins; or if they were taking sulfonamides, since PABA may interfere with the action of these. The nurse then provided information about the purpose of the study, the procedures involved, supplied all necessary equipment, and made arrangements for collection of urine samples and the timing of the second visit.

Three spot urine samples and a 24-hour sample were collected, and despatched to a laboratory for analysis.

Protocol

Respondents collected their first spot urine samples as follows:

Day 1:

- Respondents collected their first spot sample – last urine pass of the evening

Day 2:

- Respondents collected their second spot sample – first urine pass of the morning
- All urine passed after the second spot sample were collected for the 24 hour sample

Day 3:

- The 24 hour collection continued until the first urine pass of the morning.

The third spot sample was collected before or during the second nurse visit, which usually took place on day 3.

Respondents were given detailed written instructions (see Appendix A), and were provided with the following equipment:

- a 400 ml plastic beaker
- one or two 2 litre screw capped plastic bottles – the collection containers
- a safety pin (as an aide memoire)
- 2 plastic bags for carrying the equipment
- a blister pack of three PABA tablets
- three plastic syringes with small push-on caps and plastic extension tubes
- three 100ml disposable beakers
- an instruction card showing in pictures how to collect the spot samples

For the spot samples, respondents they were instructed to pass mid-flow urine into a small beaker, and to take a sample from the beaker using the plastic syringe supplied.

For the 24-hour collection, they were instructed to pass urine into the 400ml beaker, then pour it into the large collection container. Plastic bags were provided to carry the equipment if respondents were not at home for some of the collection period.

Three PABA tablets were provided, with the instruction that these should be taken at approximately even intervals throughout the 24-hour collection period, ideally with or after meals. Nurses wrote the suggested times for taking the tablets on a diary left with respondents, and they were asked to record the time that they actually took them, as well as the start and finish times of their collection, any missed urine passes, and any medication taken during the collection.

Typically the second nurse visit took place within one day from the 24-hour urine collection. The nurse checked the diary to ensure that PABA tablets had been taken, and took an aliquot from the 24-hour sample during the second visit. Samples were not accepted if all three PABA tablets had not been taken.

After the second nurse visit, all samples were labelled and despatched to the Doctor's Laboratory, London, where the analyses of sodium, potassium and chloride were carried out by ion-selective electrode methods on the Roche/Hitachi systems, using specially prepared aqueous solutions containing a growth-inhibitory preservative. Following lab analysis, an aliquot of the 24-hour sample was sent to the MRC Dunn Human Nutrition laboratory (Cambridge) for an assessment of the completeness of the 24-hour collection. Completeness was assessed using the para-amino-benzoic acid (PABA) recovery method.^{iv} In brief, the method involved administering three 80mg PABA tablets over the 24-hour sample collection period. Completion of the collection was determined by the proportion of PABA recovered in the 24-hour sample.

5 Weighting

The Primary Sampling Units (PSUs) selected for the this study were clusters of those used in the Scottish Health Survey (SHS). The PSUs drawn for the SHS were drawn disproportionately to ensure that all Health Boards were represented and that minimum sample sizes were achieved in each. Thus certain Health Board areas had a higher chance of being included in the survey. For analysis of the SHS, a set of selection weights were generated to adjust for this over-sampling in some areas. These PSU-level selection weights were also applied to the Scottish Sodium Study sample so that the results were nationally representative. In addition the households in the sample re-contacted from the SHS were also given the SHS household non-response weight.

At each household up to two adults aged 19-64 were selected at random and invited to participate in the study. Selecting two persons per household means people living in households with three or more eligible people are under-represented in the sample. A

person selection weight is required; this is the inverse of the person selection probability and is equal to 1 where there are fewer than 3 persons per household. Where there are 3 or more eligible persons the weight is equal to the number of eligible adults in the household divided by 2.

Calibration weighting was applied to the weights already described, to create a new set of weights that adjusted the profile of the achieved sample to match the population of persons aged 19-64 in terms of age and sex. The population figures were taken from the 2005 mid-year population estimates generated by General Register Office for Scotland (GROS).

Table C: Selection and final weights

Age by sex	Respondents weighted by selection weights	Respondents weighted by final weights	Population based on GROS 2005 mid-year population estimates
	%	%	%
Male 19-29	3.6	11.2	11.2
Male 30-39	7.6	11.0	11.0
Male 40-49	11.2	12.0	12.0
Male 50-59	14.5	10.5	10.5
Male 60-64	6.5	4.2	4.2
Female 19-29	5.3	11.1	11.1
Female 30-39	11.3	11.9	11.9
Female 40-49	16.8	12.7	12.7
Female 50-59	16.7	10.9	10.9
Female 60-64	6.5	4.5	4.5
Base (unweighted)	574	574	3,137,000

6 Statistical analysis

24-hour urine samples

The aims of the 24-hour urine analysis were:

- a) to provide estimates and distribution of the average 24-hour Sodium concentrations, and
- b) to estimate the mean 24-hour salt intake.

As in the 2001 National Diet and Nutrition Survey, salt intake was estimated as 1 g salt =17.1 mmol of sodium.

Mann-Whitney tests were used to test for sodium and salt differences between men and women, and Kruskal-Wallis tests examined differences among age groups. No statistical

tests by sex within age groups were performed due to the relatively small sample size of each age group.

Analysis of para-amino-benzoic acid (PABA)

Twenty-four-hour urine collection containing between 85% and 110% of the PABA marker were considered complete. Urine samples with a PABA recovery under 70% were excluded as incomplete. Urine samples with over 110% of PABA recovery were considered high and they were re-analysed using the HPLC method.¹ HPLC-analysed samples were also included if the PABA recovery was between 85% and 110% of PABA recovery. Urine samples with 70-84% PABA recovery (HPLC or direct methods) were included after adjustments.^v The adjustment was made by using the following equations:

Corrected 24-hour Sodium= Sodium*(93 / Percentage PABA recovery)

Corrected 24-hour Potassium=Potassium *(93 / Percentage PABA recovery)

Corrected 24-hour Creatinine= Creatinine *(93 / Percentage PABA recovery)

These adjusted values were used for all results presented in this report.

Spot urine samples

The aims of the spot urine samples analysis were:

a) to determine the magnitude and statistical significance of the correlation between the Na and K concentrations (adjusted for creatinine) of each individual spot samples and that of the 24-hour sample (adjusted for creatinine). As urinary sodium and urinary potassium did not show a normal distribution, non-parametric methods were used to analyse the data. Spearman correlation coefficient was used to measure the correlation between the three individual spot samples and the 24-hour sample.

b) to assess to what extent each spot urine sample could differentiate between different levels of Na and K in the 24-hour sample. This was done by plotting the quintiles of the 24-hour Na and K (adjusted for creatinine) against the corresponding Na and K means of the three spot samples (adjusted for creatinine as well).

Analysis of sodium, potassium and creatinine

While large diurnal variations in excretion of Na, K and water occur, creatinine (Cre) excretion is relatively constant from day to day (coefficient of variation is approximately 11%)^{vi}. Hence the ratio of Na and K excretion to the creatinine excretion can be used to standardise spot urine samples for overall dietary electrolyte intakes. Creatinine correction has been used in other studies that compared sodium and potassium concentrations between spot and 24-hour collections.^{vii, viii}

The spot and 24-hour ratios of Na/Cre and K/Cre were the main outcomes of the spot urine analyses.

7 Results

There were 564 useable samples processed in the laboratory. In total, 19% of men (n=47) and 23% of women (n=75) were excluded from this analysis because of incomplete urine samples. In terms of age, the included sample was not significantly different from the sample excluded from the analysis (mean age 48.1 in the included sample and 48.3 years in the excluded sample in men, and 47.3 and 44.1 respectively in women).

Results are shown separately by sex and age group. Tables 1 and 2 provide means of urinary sodium levels and distribution, and Tables 3 and 4 show mean salt intake and distribution. The mean salt intake was 9.1 g/day, with a mean of 10.6 g/day among men and 7.6 g/day among women. Younger men (aged 19-64) had the highest mean daily intake at 12.0 g/day. Overall, threequarters of the population had a daily intake higher than the recommended 6 g/day (85% of men, 65% of women).

Tables 5 and 6 present the descriptive statistics for Na, K and creatinine concentrations in the three spot samples and the 24-hour urine sample by sex and age group, respectively.

Tables 7 and 8 show the non-parametric correlation coefficients of the Na/Cre and K/Cre ratios for each of the spots and the 24-hour sample. The correlations are all statistically significant at the $p < 0.001$ level, with the exception of the Na/Cre ratio for Spot 3 among respondents aged 55-64, which was statistically significant at the $p < 0.005$ level. The correlations were slightly stronger for women in most cases, though there was no clear pattern according to age.

Tables 9 and 10 show the mean and median Na/Cre and K/Cre ratios of the spot samples against quintiles of the 24-hour quintiles, and the means have been plotted in charts 1-4.

The main aim of comparing the spot and 24-hour samples in this study was to establish the extent to which the information gained from spot samples reflects the 'true' levels of the electrolyte in the population as measured by the 24-hour sample. Results from spot samples may be considered valid, provided that the means for the Na/Cre and K/Cre from the spot urine samples follow the same patterns as the 24-hour samples for the variables of interest. This analysis showed significant though not very strong correlations between the spot samples and the 24-hour samples.

8 Discussion

The three spot samples correlated significantly with the 24-hour sample in terms of Na and K concentrations adjusted for creatinine. Overall, these results support the ability of a single spot urine sample to differentiate between subgroups of the population in a similar way to the 24-hour sample.

Correlation coefficients, although relatively weak, are of similar magnitude to those reported in other studies that have compared Na excretion in spot urine and 24-hour urine samples. For example, a study in Japan^{ix} that compared Na levels of a single spot and a 24-hour sample reported a correlation coefficient of 0.47. Higher correlation coefficients

for Na ($r=0.77$) and K ($r=0.81$) have been reported in a study^{viii} that compared the averages of several spot samples collected over a period of 3 days and the average of three 24-hour samples. However, no direct comparisons can be made with the results reported here due to substantial differences in the analytical approach.

The use of the PABA method allowed the exclusion of those cases that did not comply adequately with the 24-hour collection. In addition, the PABA method allowed the correction of a number of cases that missed a few urine collections during the 24-hour period. It is worth mentioning that the inclusion or exclusion of these “correctable” cases did not have a measurable effect on the magnitude of the reported correlations.

The results of this study confirm those of an earlier study, and suggest that spot urine samples may be used to monitor trends in dietary sodium and to compare subgroups of the population, even if they do not exactly replicate the electrolyte levels of 24-hour samples. In this context, the ability to discriminate between different subgroups is important. The fact that the means for the Na/Cre and K/Cre from the spot urine samples followed the same patterns as the 24-hour samples suggest that the spot sample could differentiate between subgroups of the population in a similar way to the 24-hour sample.

TABLES

- 1 Mean urinary sodium (mmol/24hr), by sex and age
- 2 Percentage distribution of total urinary sodium (mmol/24hr), by sex and age
- 3 Mean estimated salt (g/day), by sex and age
- 4 Percentage distribution of estimated salt intake (g/day), by sex and age
- 5 Descriptive statistics of sodium, potassium, and creatinine concentrations (mmol/L), by sex
- 6 Descriptive statistics of sodium, potassium, and creatinine concentrations (mmol/L), by age
- 7 Nonparametric Correlations (Spearman) between spot and 24-hour samples (adjusted for Creatinine), by sex
- 8 Nonparametric Correlations (Spearman) between spot and 24-hour samples (adjusted for Creatinine), by age
- 9 Mean and median sodium/ creatinine (mmol/L) of spot urine samples by 24-hour sodium/ creatinine quintile
- 10 Mean and median potassium/ creatinine ratio (mmol/L) of spot urine samples by 24-hour potassium/ creatinine quintile

Table 1 Mean urinary sodium (mmol/24hr), by sex and age

Corrected 24hr Sodium (mmol/24hr)

	Age group				Total
	19-24	25-34	35-49	50-65	
Men					
Mean	205	166	193	163	181
Standard Error	13	13	8	8	5
Standard Deviation	69	81	71	62	71
Lower 2.5 centile	110	72	77	67	72
Top 2.5 centile	-	-	338	330	338
Median	215	140	198	166	181
Women					
Mean	128	139	131	123	130
Standard Error	19	12	6	6	4
Standard Deviation	80	69	59	47	59
Lower 2.5 centile	52	34	33	38	34
Top 2.5 centile	-	-	258	252	271
Median	92	128	122	113	117
All					
Mean	175	153	162	143	156
Standard Error	12	9	5	5	3
Standard Deviation	82	76	72	59	71
Lower 2.5 centile	52	34	39	52	52
Top 2.5 centile	320	365	335	281	330
Median	185	140	157	136	144
<i>Bases (weighted)</i>					
<i>Men</i>	29	37	89	69	224
<i>Women</i>	19	33	87	66	206
<i>Bases (unweighted)</i>					
<i>Men</i>	5	18	76	96	195
<i>Women</i>	7	22	105	113	247

Table 2 Percentage distribution of total urinary sodium (mmol/24hr), by sex and age

<i>mmol/24hr</i>	Age group				Total
	19-24	25-34	35-49	50-65	
	%	%	%	%	%
Men					
Under 60	-	-	1	1	1
Under 90	-	13	6	9	7
Under 120	26	31	20	26	31
Under 150	26	55	25	26	55
Under 180	26	64	41	60	49
Under 210	40	80	64	82	69
Under 270	86	87	86	95	89
Women					
Under 60	34	10	7	5	9
Under 90	40	20	20	22	23
Under 120	53	44	49	58	51
Under 150	63	70	69	79	72
Under 180	81	75	81	87	82
Under 210	81	92	88	93	90
Under 270	81	96	99	100	97
All					
Under 60	14	5	4	3	5
Under 90	16	16	13	16	15
Under 120	37	37	34	42	38
Under 150	41	62	47	60	53
Under 180	48	69	61	74	65
Under 210	56	86	76	87	79
Under 270	84	91	92	97	93
<i>Bases weighted)</i>					
<i>Men</i>	29	37	89	69	224
<i>Women</i>	19	33	87	66	206
<i>Bases (unweighted)</i>					
<i>Men</i>	5	18	76	96	195
<i>Women</i>	7	22	105	113	247

Table 3 Mean estimated salt (g/day), by sex and age

<i>g/day</i>	Age group				
	19-24	25-34	35-49	50-65	Total
Men					
Mean	12.0	9.7	11.3	9.6	10.6
Standard Error	0.8	0.8	0.4	0.4	0.3
Standard Deviation	4.1	4.7	4.2	3.6	4.2
Lower 2.5 centile	6.5	4.2	4.5	3.9	4.2
Top 2.5 centile			19.8	19.3	19.8
Median	12.5	8.2	11.6	9.7	10.6
Women					
Mean	7.5	8.1	7.7	7.2	7.6
Standard Error	1.1	0.7	0.4	0.3	0.2
Standard Deviation	4.7	4.0	3.5	2.7	3.5
Lower 2.5 centile	3.1	2.0	1.9	2.2	2.0
Top 2.5 centile			15.1	14.7	15.9
Median	5.4	7.5	7.1	6.6	6.9
All					
Mean	10.2	9.0	9.5	8.4	9.1
Standard Error	0.7	0.5	0.3	0.3	0.2
Standard Deviation	4.8	4.4	4.2	3.4	4.1
Lower 2.5 centile	3.1	2.0	2.3	3.0	3.0
Top 2.5 centile	18.7	21.3	19.6	16.5	19.3
Median	10.8	8.2	9.2	8.0	8.4
<i>Bases (weighted)</i>					
<i>Men</i>	29	37	89	69	224
<i>Women</i>	19	33	87	66	206
<i>Bases (unweighted)</i>					
<i>Men</i>	5	18	76	96	195
<i>Women</i>	7	22	105	113	247

**Table 4 Percentage distribution of estimated salt intake (g/day),
by sex and age**

<i>g/day</i>	Age group				Total
	19-24	25-34	35-49	50-65	
	%	%	%	%	%
Men					
3 or Less	-	-	-	1	0
6 or Less	-	22	13	19	15
9 or Less	26	64	25	45	38
12 or Less	40	80	61	82	68
15 or Less	86	87	84	94	88
18 or Less	86	92	89	98	92
<i>Over 6g</i>	100	78	87	81	85
Women					
3 or Less	-	6	6	4	5
6 or Less	53	29	37	32	35
9 or Less	81	70	69	79	74
12 or Less	81	92	88	93	90
15 or Less	81	96	96	99	96
18 or Less	100	96	99	100	99
<i>Over 6g</i>	47	71	63	68	65
All					
3 or Less	-	3	3	2	2
6 or Less	21	25	25	25	25
9 or Less	48	67	47	62	55
12 or Less	56	86	74	87	78
15 or Less	84	91	90	96	92
18 or Less	91	94	94	99	95
<i>Over 6g</i>	79	75	75	75	75
Bases (weighted)					
<i>Men</i>	29	37	89	69	224
<i>Women</i>	19	33	87	66	206
Bases (unweighted)					
<i>Men</i>	5	18	76	96	195
<i>Women</i>	7	22	105	113	247

Table 5: Descriptive statistics of sodium, potassium, and creatinine concentrations (mmol/L), by sex															
	<i>Mean</i>			<i>Standard Error</i>			<i>Standard Deviation</i>			<i>Minimum</i>			<i>Maximum</i>		
	Men	Women	All	Men	Women	All	Men	Women	All	Men	Women	All	Men	Women	All
Sodium															
Spot 1	107.3	99.2	103.4	4.23	4.05	2.94	63.34	57.99	60.91	9.0	9.0	9.0	289.0	256.0	289.0
Spot 2	99.2	97.1	98.2	3.06	3.54	2.32	45.75	50.78	48.18	10.0	10.0	10.0	240.0	260.0	260.0
Spot 3	112.7	91.9	102.7	3.79	3.69	2.70	56.46	52.83	48.18	17.0	9.0	9.0	265.0	225.0	265.0
24-hour sample	181.0	129.5	156.3	4.77	4.13	3.41	71.37	59.26	70.63	51.1	32.1	32.1	447.1	391.2	447.1
Potassium															
Spot 1	46.6	38.8	42.9	2.15	1.97	1.47	32.09	28.26	30.53	1.2	1.8	1.2	128.3	160.1	160.1
Spot 2	45.3	42.9	44.2	1.44	1.62	1.08	21.55	23.19	22.36	2.0	4.5	2.0	105.2	144.4	144.4
Spot 3	69.1	61.2	65.3	2.64	2.66	1.88	39.27	38.01	38.83	6.7	2.5	2.5	210.6	249.5	249.5
24-hr sample	84.5	66.1	75.7	1.70	1.41	1.20	25.44	20.21	24.81	16.3	19.8	16.3	153.6	165.2	165.2
Creatinine															
Spot 1	11.7	8.3	10.1	0.46	0.37	0.31	6.89	5.26	6.39	0.9	0.5	0.5	37.5	30.8	37.5
Spot 2	14.6	10.9	12.8	0.50	0.41	0.34	7.51	5.84	6.99	1.3	1.3	1.3	39.4	26.1	39.4
Spot 3	12.3	9.6	11.0	0.32	0.42	0.29	5.85	6.03	6.09	1.2	0.7	0.7	33.2	37.5	37.5
24-hour sample	17.9	10.7	14.4	0.32	0.22	0.26	4.78	3.11	5.44	6.3	3.4	3.4	35.6	22.2	35.6
Base:	Weighted	Un-weighted													
Men	224	195													
Women	206	247													
All	430	442													

Table 6: Descriptive statistics of sodium, potassium, and creatinine concentrations (mmol/L), by age

	Mean				Standard Error				Standard Deviation				Minimum				Maximum			
	19-24	25-34	35-49	50-64	19-24	25-34	35-49	50-64	19-24	25-34	35-49	50-64	19-24	25-34	35-49	50-64	19-24	25-34	35-49	50-64
Sodium																				
Spot 1	103.8	97.1	110.1	98.0	10.06	7.81	4.65	4.61	69.47	65.66	61.62	53.57	12.0	13.0	9.0	9.0	188.0	248.0	289.0	289.0
Spot 2	109.8	99.4	97.9	93.9	9.29	5.75	3.51	3.71	64.18	48.34	46.71	43.05	23.0	15.0	10.0	14.0	227.0	207.0	260.0	244.0
Spot 3	137.9	116.7	95.6	92.1	10.31	6.06	3.97	4.28	71.23	50.92	52.67	49.31	25.0	20.0	9.0	9.0	265.0	211.0	251.0	248.0
24-hr sample	175.0	153.3	162.4	143.5	11.91	9.05	5.44	5.04	82.23	76.03	72.25	58.51	52.4	33.5	32.1	32.5	320.0	378.0	391.2	447.1
Potassium																				
Spot 1	41.3	38.8	44.4	43.7	5.46	3.35	2.25	2.59	37.73	28.16	29.76	30.03	3.9	2.0	1.2	2.8	160.1	101.0	144.9	128.3
Spot 2	62.4	45.5	42.3	39.5	3.12	2.47	1.59	1.89	21.51	20.75	21.14	22.00	10.6	16.7	2.0	4.6	99.2	124.4	119.7	144.4
Spot 3	79.4	72.0	61.1	62.3	6.30	4.39	3.01	3.06	43.48	36.90	39.90	35.25	8.6	9.2	2.5	4.4	164.3	186.4	249.5	206.3
24-hr sample	69.8	75.0	78.2	74.8	2.48	2.65	2.17	1.92	17.11	22.30	28.83	22.35	26.0	21.3	16.3	19.8	98.2	130.4	165.2	144.0
Creatinine																				
Spot 1	9.2	10.1	10.8	9.5	0.82	0.83	0.48	0.54	5.63	6.98	6.40	6.30	2.2	1.0	0.5	0.7	17.9	25.4	30.5	37.5
Spot 2	19.5	13.2	12.8	10.3	1.44	0.74	0.48	0.45	9.95	6.22	6.33	5.23	2.3	2.5	1.3	1.3	39.4	27.6	31.7	26.9
Spot 3	11.0	12.8	10.9	10.3	0.59	0.75	0.49	0.50	4.06	6.32	6.55	5.80	4.5	2.3	0.7	0.9	17.4	25.2	37.5	28.8
24-hour sample	15.7	14.3	15.1	13.2	0.82	0.61	0.43	0.42	5.69	5.08	5.75	4.89	5.0	4.7	4.0	3.4	24.4	23.7	35.6	34.9
Bases:	wt	unwt																		
19-24	48	12																		
25-34	71	40																		
35-49	177	181																		
50-64	135	209																		

Table 7. Nonparametric Correlations (Spearman) between spot and 24-hour samples (adjusted for Creatinine), by sex

	24-hour sodium (Na) / 24-hour creatinine (Cre)								
	Men			Women			All		
	Coeff.	Sig. (2-tailed)	N	Coeff.	Sig. (2-tailed)	N	Coeff.	Sig. (2-tailed)	N
Spot 1 sodium (Na) / creatinine (Cre)	0.29	0.000	237	0.47	0.000	220	0.42	0.000	457
Spot 2 sodium (Na) / creatinine (Cre)	0.33	0.000	237	0.59	0.000	220	0.50	0.000	457
Spot 3 sodium (Na) / creatinine (Cre)	0.56	0.000	237	0.40	0.000	220	0.48	0.000	457

	24-hour potassium (K) / 24-hour creatinine (Cre)								
	Men			Women			All		
	Coeff.	Sig. (2-tailed)	N	Coeff.	Sig. (2-tailed)	N	Coeff.	Sig. (2-tailed)	N
Spot 1 potassium (K) / creatinine (Cre)	0.36	0.000	237	0.47	0.000	220	0.47	0.000	457
Spot 2 potassium (K) / creatinine (Cre)	0.36	0.000	237	0.51	0.000	220	0.49	0.000	457
Spot 3 potassium (K) / creatinine (Cre)	0.35	0.000	237	0.46	0.000	220	0.44	0.000	457

Table 8. Nonparametric Correlations (Spearman) between spot and 24-hour samples (adjusted for Creatinine), by age

	24-hour sodium (Na) / 24-hour creatinine (Cre)								
	16-39 yrs			40-54 yrs			55-64 yrs		
	Coeff.	Sig. (2-tailed)	N	Coeff.	Sig. (2-tailed)	N	Coeff.	Sig. (2-tailed)	N
Spot 1 sodium (Na) / creatinine (Cre)	0.42	0.000	185	0.45	0.000	155	0.38	0.000	117
Spot 2 sodium (Na) / creatinine (Cre)	0.47	0.000	185	0.53	0.000	155	0.54	0.000	117
Spot 3 sodium (Na) / creatinine (Cre)	0.52	0.000	185	0.54	0.000	155	0.28	0.000	117

	24-hour potassium (K) / 24-hour creatinine (Cre)								
	16-39 yrs			40-54 yrs			55-64 yrs		
	Coeff.	Sig. (2-tailed)	N	Coeff.	Sig. (2-tailed)	N	Coeff.	Sig. (2-tailed)	N
Spot 1 potassium (K) / creatinine (Cre)	0.51	0.000	185	0.39	0.000	155	0.46	0.000	117
Spot 2 potassium (K) / creatinine (Cre)	0.43	0.000	185	0.52	0.000	155	0.53	0.000	117
Spot 3 potassium (K) / creatinine (Cre)	0.33	0.000	185	0.46	0.000	155	0.54	0.000	117

Table 9 Mean and median sodium/ creatinine (mmol/L) of spot urine samples by 24-hour sodium/ creatinine quintile

Men	<i>NA/ Cre quintiles of 24-hour Sodium</i>				
	Bottom	2nd	3rd	4th	Top
Ratio Na/Cre for spot 1:					
Mean	7.8	10.4	10.8	11.3	11.9
Median	6.9	9.9	10.1	9.1	10.5
Standard Error	0.5	0.7	0.8	0.9	0.9
Standard Deviation	3.6	4.6	5.9	5.2	6.2
Ratio Na/Cre for spot 2:					
Mean	6.4	8.5	7.4	9.0	10.4
Median	5.4	7.1	6.9	7.6	8.8
Standard Error	0.8	0.6	0.7	0.7	0.9
Standard Deviation	5.4	3.7	5.0	4.1	6.6
Ratio Na/Cre for spot 3:					
Mean	6.9	7.9	11.1	10.6	15.5
Median	5.9	6.4	9.8	10.2	14.3
Standard Error	0.5	0.8	0.6	0.8	0.9
Standard Deviation	3.1	5.3	4.4	5.0	6.2
Women					
Ratio Na/Cre for spot 1:					
Mean	8.9	11.8	14.4	16.4	18.5
Median	8.3	9.7	11.9	13.6	15.5
Standard Error	0.7	0.9	1.2	1.3	1.3
Standard Deviation	4.4	6.1	7.9	8.3	7.9
Ratio Na/Cre for spot 2:					
Mean	5.4	9.2	10.0	12.5	16.3
Median	5.4	8.1	9.8	11.3	14.2
Standard Error	0.5	0.8	0.6	0.9	1.3
Standard Deviation	2.8	5.5	4.4	5.8	7.6
Ratio Na/Cre for spot 3:					
Mean	7.9	10.1	9.9	13.1	19.1
Median	6.4	9.5	7.6	12.8	16.2
Standard Error	0.8	0.7	0.8	1.0	1.8
Standard Deviation	5.3	4.8	5.4	6.2	11.1

Table 10 Mean and median potassium/ creatinine ratio (mmol/L) of spot urine samples by 24-hour potassium/ creatinine quintile

Men	<i>K/ Cre quintiles of 24-hour Sodium</i>				
	Bottom	2nd	3rd	4th	Top
Ratio K/Cre for spot 1:					
Mean	2.9	3.4	3.7	4.5	6.0
Median	2.4	3.0	3.1	3.8	5.3
Standard Error	0.2	0.2	0.3	0.4	0.5
Standard Deviation	1.4	1.5	2.3	2.8	3.1
Ratio K/Cre for spot 2:					
Mean	2.6	3.3	3.8	3.6	4.6
Median	2.3	3.1	3.3	3.2	3.8
Standard Error	0.1	0.2	0.2	0.3	0.4
Standard Deviation	0.9	1.3	1.7	1.7	2.8
Ratio K/Cre for spot 3:					
Mean	4.5	5.4	6.1	7.0	6.8
Median	3.5	5.1	6.1	7.0	6.4
Standard Error	0.3	0.4	0.3	0.6	0.4
Standard Deviation	2.3	2.3	2.3	3.7	2.5
Women					
Ratio K/Cre for spot 1:					
Mean	3.6	4.0	5.1	5.9	7.0
Median	3.3	3.4	4.6	5.4	6.4
Standard Error	0.3	0.2	0.4	0.4	0.6
Standard Deviation	1.8	1.7	2.5	2.8	3.9
Ratio K/Cre for spot 2:					
Mean	3.3	4.0	4.2	4.5	6.0
Median	2.9	3.4	3.7	4.0	5.4
Standard Error	0.2	0.3	0.3	0.3	0.3
Standard Deviation	1.4	2.1	1.6	1.7	2.0
Ratio K/Cre for spot 3:					
Mean	4.9	6.5	6.7	7.4	10.3
Median	4.6	6.2	6.0	7.5	9.3
Standard Error	0.3	0.4	0.5	0.4	0.8
Standard Deviation	2.2	2.8	3.1	2.7	4.9

Chart 1: Men's mean sodium (Na) concentrations (adjusted for creatinine) in the three spot samples by 24-hour Na quintile

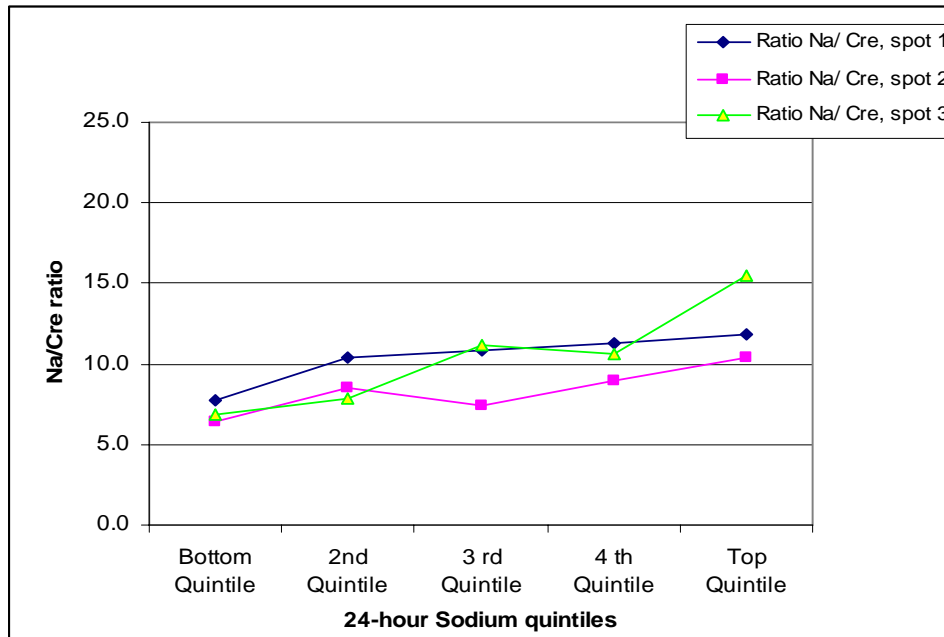


Chart 2: Women's mean sodium (Na) concentrations (adjusted for creatinine) in the three spot samples by 24-hour Na quintile.

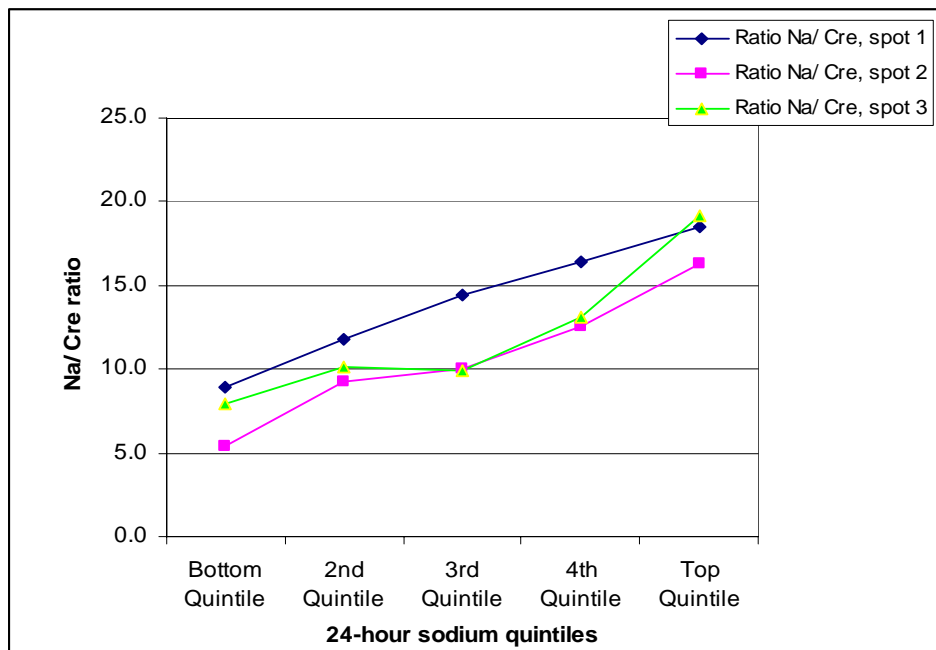


Chart 3: Men's mean potassium (K) concentrations (adjusted for creatinine) in the three spot samples by 24-hour Na quintile.

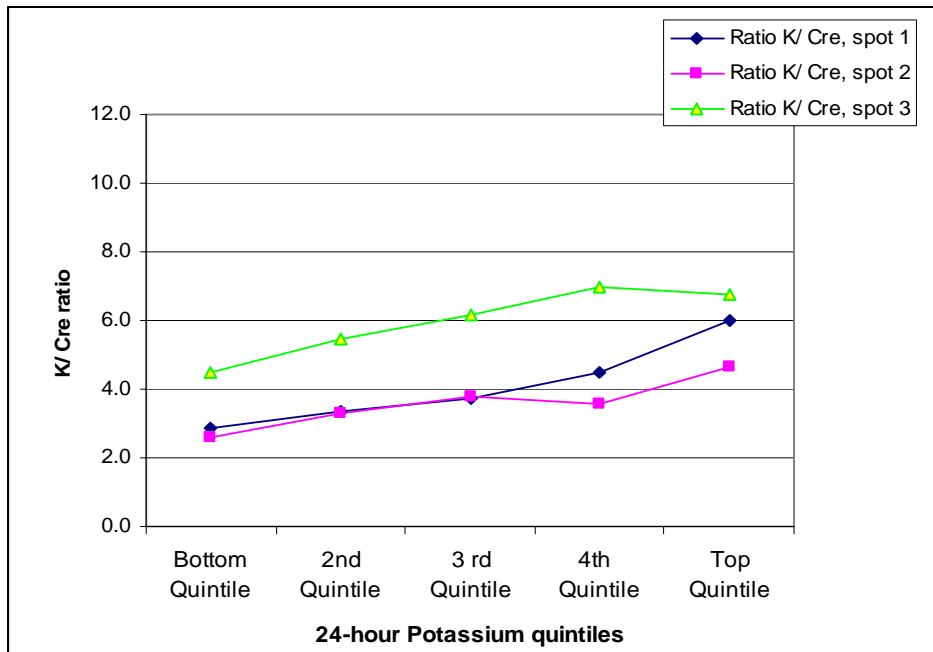
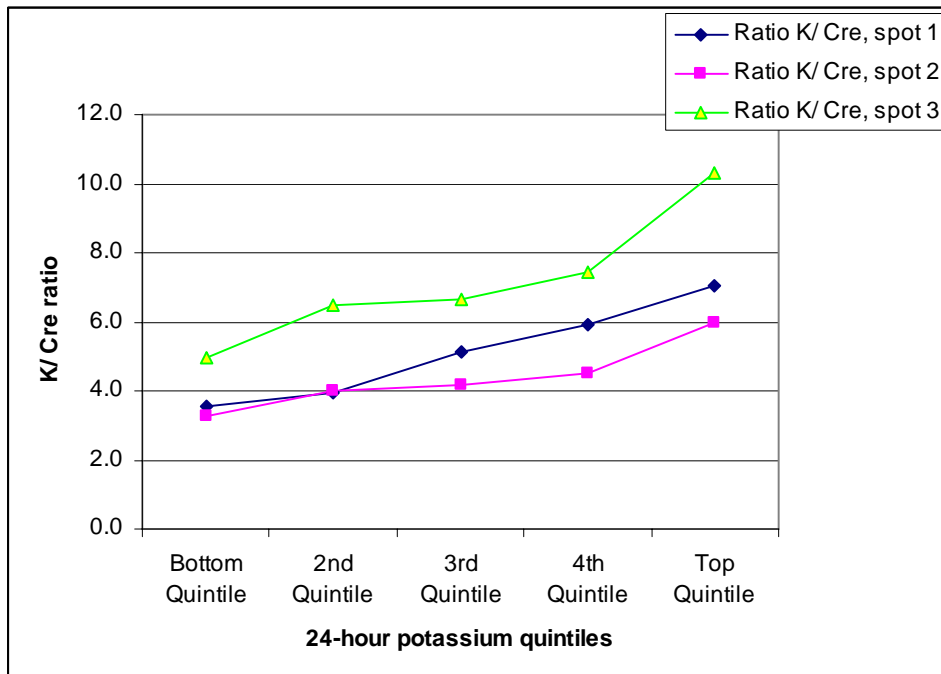


Figure 2b: Women's mean potassium (K) concentrations (adjusted for creatinine) in the three spot samples by 24-hour Na quintile.



APPENDIX A: RESPONDENT INSTRUCTIONS

P7014
Blue Team

A Study of Salt Levels in People's Diet

Instructions

Thank you for agreeing to take part in this study. Some information about this study and instructions for collecting urine samples are given here.

Introduction

Levels of salt in the diet can have an effect on health. It is possible to measure levels of salt in the diet by measuring salt levels in urine. Urine samples can be collected on the spot, e.g., during a visit by the nurse, or can be collected over a longer period of time.

Salt levels in urine vary due to salt in the diet, the amount a person has drunk and time of day. These variations can be reduced by taking a sample from urine collected over a 24-hour period.

In this study we need to compare salt levels in 'spot' urine samples, such as those collected during a nurse visit, with salt levels in a 24 hour sample of urine. Each person who takes part in this study will need to provide three samples of urine and to collect their urine over a 24 hour period. The urine samples will be only tested for salt levels. We will not test the sample for drugs or viruses.

The urine samples will be collected at times agreed with the nurse. The pattern of collections will be as follows:

- 1st spot sample: Day 1 (final urine pass of evening):**
- 2nd spot sample Day 2 (first urine pass of morning):**
- 24 hour sample Day 2 (from second urine pass) until Day 3 (first morning urine pass included in collection)**
- 3rd spot sample Day 3 or 4 (during or immediately before the second nurse visit)**

Details of the equipment and instructions on how to collect the 24 hour and spot urine samples are shown overleaf.

The 24 hour urine sample

- *The equipment provided for the 24 hour collection*

The nurse will give you the following equipment:

1. a 400 ml plastic beaker
2. Two 2 litre screw capped plastic bottles (the collection containers)
3. a safety pin
4. 2 plastic bags for carrying the equipment
5. a blister pack of three PABA tablets.

NOTE: The 2-litre plastic bottle – the collection container - contains a boric acid preservative. This could cause skin or eye-irritations by contact or could cause stomach upset if swallowed. There is a warning label on the bottle but please be sure to keep it out of the reach of young children.

- *When to collect the 24 hour sample*

The sample should be collected during the agreed 24-hour period. The nurse will help you to choose a day on which you would like to make the 24-hour urine collection. You may prefer to choose a day when you will be mostly at home or away for only a short time. If you are female, you may prefer not to make your collection during your period.

Please start your collection from the second morning pass and collect all daytime and night-time urine until the first morning pass the following day. During this time, use the safety pin provided to pin your underclothes to your outer garments or nightwear to remind you to collect your urine.

- *Collecting your urine for the 24 hour sample*

Please follow these instructions during the 24-hour collection period.

1. Pass all urine directly into the **400ml plastic beaker**.
2. Pour urine from the beaker into the **collection container**
3. If you need to open your bowels, always remember to pass urine first **before** you pass a stool.

- *The PABA tablets*

Three tablets are taken over the 24 hours. An information leaflet will be provided, along with a diary that tells you when to take these. It is important that you take these so that we can measure how complete the urine sample is.

- *What happens if you miss any collections?*

If during the 24 hour collection a sample is missed for any reason, such as because of a bowel motion, we would like you to record this on the **24 hour urine study diary**.

- *The 24 hour urine study diary*

The diary is used to record important information about the samples. The nurse will fill in some details including the agreed date and time for the 24-hour collection and when to take the PABA tablets. We need you to write down:

- date and time you collected the spot samples
- date and time of any missed collections
- all medicines or vitamins you have taken during the 24 hours

If you have any questions about the 24-hour sample please speak to the nurse.

The spot urine samples

'Spot' urine samples are those collected during a single visit. You will need to collect three spot samples during the study, two before the 24-hour sample and one after.

- *Equipment provided to collect the spot samples*

The nurse will give you the following:

1. three plastic syringes with small push-on caps and plastic extension tubes.
2. three 100ml disposable beakers.
3. an instructions card that shows in pictures how to collect the samples

- *When to collect the spot samples*

The 1st spot sample is collected the evening before the 24-hour urine collection – this should be the last void before you go to bed.

The 2nd spot sample is collected first thing in the morning, before you start the 24-hour collection.

The 3rd spot sample is collected when the nurse returns to collect the samples, or just before the visit if you need to visit the bathroom before the nurse arrives.

- *Collecting your urine for the spot sample*

1. Collect a sample of your urine using the beaker provided – the sample should be collected midflow. Use the syringe to take a sample of this urine from the beaker. The pictures on the **instructions card** show you how to use the syringe.
2. Remove the small push cap from the syringe (Picture 2). Do **not** remove the larger screw-cap where the preservative is kept.
3. Push the extension tube tightly onto the exposed syringe nozzle.
4. Put the end of the extension tube into the urine in the beaker and pull back the syringe plunger to fill the syringe. Make sure that you pull the plunger right to the end of the tube.
5. Remove the extension tube and replace the push cap. Push this on firmly to make sure no urine leaks out.
6. Break off the plunger stalk by snapping it off.

After this you can dispose of the urine that is left in the beaker, pouring it down the toilet. The beaker and extension tubes should be rinsed and disposed of along with your usual household waste.

- *Labelling and storage*

The nurse will stick a green label onto each of the urine syringes. The nurse will fill in some details but, when you have collected the sample, you will need to write in:

- the tube number - '1' for the 1st spot, '2' for the second
- your initials

The syringe should be stored at room temperature, until the nurse returns to collect the samples.

If you have any questions about the spot urine samples please speak to the nurse.

Urine Sample Syringe Instructions

P7014

1. Collect your sample in the disposable pot

2. Remove the small push cap.



3. Push the extension tube on the syringe nozzle.



4. Pull back the syringe plunger to fill the syringe.



5. Remove the extension tube.



5. Replace the cap.



6. Pull the syringe plunger until it clicks...



...and break off the stalk.

References:

- ⁱSacks FM, Svetkey LP, Vollmer WM et al. Effects on blood pressure of reduced dietary sodium and the Dietary Approaches to Stop Hypertension (DASH) diet. DASH-Sodium Collaborative Research Group. *New Engl J Med* 2001;344:3
- ⁱⁱ Department of Health (1991). Dietary Reference Values for Food, Energy and Nutrients for the United Kingdom. Report on Health and Social Subjects 41. London: The Stationery Office.
- ⁱⁱⁱ Department of Health (1994). Nutritional Aspects of Cardiovascular Disease. Report on Health and Social Subjects 46. London: The Stationery Office.
- ^{iv} Jakobsen J, Ovesen L, Fagt S, et al. Para-aminobenzoic acid used as a marker for completeness of 24 hour urine: Assessment of control limits for a specific HPLC method. *Eur J Clin Nutr* 1997; 5: 514
- ^v Johansson G, Bingham S, Vahter M. A method to compensate for incomplete 24-hour urine collections in nutritional epidemiology studies. *Public Health Nutrition*:2(4), 587-591.
- ^{vi} Bingham SA, Williams R, Cole TJ, et al. Reference values for analytes of 24-hour urine collections known to be complete. *Ann Clin Biochem* 1988; 25: 610.
- ^{vii} Brandle E, Melzer H, Gomez-Anson B, et al. Can examination of spontaneous urine samples adequately replace 24-hour-urine samples for determining excretory rate of various lithogenic and inhibitory substances in metabolic evaluation of kidney calculi patients? *Urologe A* 1996; 35(2): 136.
- ^{viii} Tanaka T, Okamura T, Miura K, et al. A simple method to estimate populational 24-hour urinary sodium and potassium excretion using a casual urine specimen. *Hum Hypertens.* 2002;16(2): 97.
- ^{ix} Kawasaki T, Ueno M, Kawazoe N, Nakamuta S, Ueda K, Omae T. Average urinary excretion of sodium in 24 hours can be estimated from a spot-urine specimen. *Jpn Circ J.* 1982; 46(9): 948.