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The Perils of Sodium

## Spot urine samples compared with 24-h urine samples for estimating changes in urinary sodium and potassium excretion in the China Salt Substitute and Stroke Study

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### Abstract

**Background**: The capacity of spot urine samples for detecting changes in population sodium and potassium excretion is unclear.

**Methods**: Changes in urinary sodium and potassium excretion, over a 6-month to 2-year interval, were measured from 24-h urine samples and estimated from spot urine samples using several published methods in 3270 Chinese. Additional estimates were made by multiplying individual spot sodium and potassium concentrations by a single estimated 24-h urine volume derived from external data.

**Results:** The measured difference in 24-h urinary excretion between intervention and control groups was -0.35 g (95% CI: -0.68 to -0.02; P = 0.039) for sodium and 0.66 g (95% CI: 0.52 to 0.80; P < 0.001) for potassium, based upon 24-h urine samples. The corresponding estimates of sodium differences for the Tanaka (-0.06 g), Kawasaki (-0.09 g), Intersalt without potassium (-0.09 g) and Intersalt with potassium (-0.14 g) equations were all smaller and identified no reduction in sodium excretion (all P > 0.10). The estimates were -0.65 g for sodium and 1.11 g for potassium using individual spot urine concentrations and an externally derived standard urine volume (both P < 0.01).

**Conclusions:** The published equations were unable to detect the differences in sodium excretion measured by 24-h urine samples. A method based upon spot urine electrolyte concentrations and a standard urine volume may offer an alternative approach to measuring differences in sodium and potassium excretion between population groups without requiring 24-h urine, but will need further investigation.

Key words: Sodium, potassium, spot urine, 24-h urine, controlled trial, random allocation

#### Key Messages

- Published equations that use spot urine samples to estimate daily sodium intake were unable to detect a difference in urinary sodium excretion between population groups.
- There were detectable differences between groups in the mean concentration of sodium and potassium in spot urine samples.
- Multiplication of the sodium concentration in spot urine by the measured 24-h urine volume provided a slight overestimate of the difference in sodium excretion compared with that determined from 24-h urine samples.
- Multiplication of the concentration in spot urine by a standard single externally derived urine volume detected differences in excretion between groups for both sodium and potassium, though it resulted in overestimates of the differences.
- Serial cross-sectional surveys of spot urine sodium and potassium concentrations, without collection of 24-h urine samples, may provide a mechanism for measuring difference in population intake levels of sodium and potassium.

#### Introduction

It is well established that excessive dietary salt (sodium chloride) intake raises blood pressure, which is a leading risk factor for stroke and other cardiovascular diseases.<sup>1–3</sup> Reducing average population salt intake has been identified as a strategy for preventing these diseases,<sup>4</sup> and member states of the World Health Organization (WHO) are seeking to reduce population salt intake by 30% by 2025.<sup>4</sup> Implementation of this strategy requires baseline and serial follow-up measurements of mean population salt intake, to document progress towards this goal. In addition, supplementation of dietary potassium has also been proved to be helpful with controlling high blood pressure.<sup>5</sup>

The usual approach to measuring mean dietary sodium and potassium intake in the population is through collection of 24-h urine samples in a representative sample.<sup>6–8</sup> However, collection of 24-h urine samples is burdensome to participants, resulting in low response rates, non-representative population samples<sup>9</sup> and poor quality urine collections. Many nations find this method expensive and difficult to use.<sup>10</sup> By comparison, spot urine samples collected at one time point during the day are easier for patients to provide and simpler for research teams to collect and analyse. Spot urine samples are already the basis for estimations of pesticide exposure<sup>11,12</sup> and iodine intake.<sup>13</sup>

Previous studies have demonstrated the capacity of several different estimating equations that incorporate

measures of spot urine sodium concentration to provide an indication of mean daily population sodium intake.<sup>14</sup> The capacity of these equations to detect differences in sodium intake over time is, however, unknown. The primary objective of this study was to understand whether spot urine can be used to replace 24-h urine in estimating sodium and potassium excretion at the population level, by comparing estimates of differences in mean 24-h sodium and potassium excretions with alternative methods based upon the urinary sodium concentration in spot urine samples.

#### Methods

This investigation was done within the China Salt Substitute and Stroke Study (SSaSS),<sup>15</sup> an ongoing large-scale cluster-randomized trial investigating the effects of a reduced sodium, added potassium salt substitute compared with usual salt on the risk of stroke (clinicaltrials.gov identifier: NCT02092090). Effects of the salt substitute on urinary sodium and potassium excretion are being monitored during the course of the study to check the integrity of the randomized intervention, through the collection of 24-h urine samples in a randomly sampled subset of villages every 12 months. Before commencement, we made plans to use the trial to test questions about the potential use of spot urine samples for making estimates of daily sodium

intake, and we collected parallel spot urine samples for this purpose. The study commenced in 2014 and is scheduled to complete in 2020. Approval for the trial, including the collection and evaluation of urine samples, was obtained from both Peking University Health Science Center Institutional Review Board and the University of Sydney Ethics Committee. All participants have provided written informed consent.

#### Participants

Participants in SSaSS are individuals 60 years old or above with uncontrolled high blood pressure, or individuals with a history of stroke, resident in 600 villages in Northern China (Liaoning, Shanxi, Hebei, Ningxia, Shaanxi). There are about 35 participants in each village involved. The SSaSS participating villages (half intervention, half control) were randomly selected for baseline (36 villages), 12month (60 villages) and 24-month (140 villages) follow-up surveys to collect urine samples, with an additional 12 villages in Shanxi province surveyed at 6 months. A randomly sampled group of 20 participants from each selected village were invited to provide urine samples, with replacement from other participants within the village to achieve that number in the event that selected individuals were unavailable. Because villages and participants were randomly selected at each time point, there are a few villages and participants that have been surveyed on multiple occasions. Participants were excluded from participation in the urine collection survey if they reported urinary incontinence, inability to collect urine samples as required, genito-urinary infection, current menstruation, pregnancy or breastfeeding.

#### Urine collection and analysis

Participants were seen face to face in their village and were first instructed to empty their bladder while providing a mid-stream urine sample into a disposable urine cup, from which 2 x 2-ml aliquots of urine were extracted (the spot urine sample). Participants were then directed to immediately commence their 24-h urine collection and the time was recorded; collection of the spot urine sample and commencement of the 24-h collection could be at any time during the day. Six 1000-ml urine containers were provided, and participants were asked to collect all voids of urine for the next 24 h. Instructions were communicated orally, with supporting printed materials. An appointment was made for the next day about 23.5 h after commencement of the 24-h urine collection, at which time the return of all six containers was sought, regardless of whether they contained urine. Participants were also asked to void one last time at that visit and the time was recorded. Enquiry about missed voids and spillage was also made. Urine from all containers was then thoroughly mixed in a single large container, urine volume was measured and 2 x 2-ml aliquots were extracted. Urine samples were not collected if participants reported diarrhoea or vomiting on the day of collection or if the urine was seen to be contaminated with blood or faeces. Samples were refrigerated and transported to a central laboratory in Beijing for analysis. Assays of sodium and potassium were done by the ion-selective electrode method, and creatinine (Cr) was assayed by the sarcosine oxidase method using the HITACHI 7600 automated biochemistry analyser.

#### Statistical analysis

The primary outcome was the difference in estimated change of mean 24-h urinary sodium excretion (g) between intervention and control groups from baseline to followup. Secondary outcomes were differences in estimated change of mean 24-h urinary potassium excretion (g), mean concentration of sodium (mmol/l), mean concentration of potassium (mmol/l) and mean urine volume (ml) from baseline to follow-up. The estimating methods that were compared were based upon:

- standard 24-h urine samples: i.e. sodium concentration in individual 24-h urine sample x volume of individual 24-h urine sample;
- published estimating equations that use spot urine samples, i.e. Intersalt,<sup>16</sup> Kawasaki<sup>17</sup> and Tanaka (see Appendix Table A1) equations.<sup>18</sup> The Intersalt equation estimates were made separately with and without inclusion of potassium concentration and in the absence of a Chinese intercept, using the extensively validated North America intercept;
- simple calculations using spot urine samples and urine volumes done in two ways: (i) individual volume spot = sodium concentration in individual spot urine sample x volume of individual 24-h urine sample; (ii) standard volume spot = sodium concentration in individual spot urine sample x 1.55 L. This was the mean urine volume recorded for a comparable Northern Chinese population in Shandong province.<sup>19</sup>

Estimates were also made using volumes 10% greater (1.71 l) and 10% lesser (1.40 l). Of note, the source of this externally derived 24-h urine volume was derived from a retracted paper. Retraction was reportedly due to an oversight made by the author not including body mass index in the multiple linear regression analysis assessing the relationship between electrolyte excretion and blood pressure in adults.<sup>20</sup> The estimate of the 24-h urine volume should

	Baseline			Follow-up				
	Intervention	Control	All	Intervention	Control	All		
Total number of participants included in analysis	272 (100%)	260 (100%)	532 (100%)	1524 (100%)	1340 (100%)	2864 (100%)		
Participants from each province, $n(\%)$								
Liaoning	0 (0%)	0 (0%)	0 (0%)	169 (11%)	135 (10%)	304 (11%)		
Shanxi	0 (0%)	0 (0%)	0 (0%)	436 (291%)	345 (26%)	781 (27%)		
Hebei	90 (33%)	84 (32%)	174 (33%)	291 (199%)	288 (21%)	579 (20%)		
Ningxia	100 (37%)	96 (37%)	196 (37%)	301 (20%)	297 (22%)	598 (21%)		
Shaanxi	82 (30%)	80 (31%)	162 (30%)	327 (21%)	275 (21%)	602 (21%)		
Male, <i>n</i> (%)	125 (45%)	131 (50%)	256 (48%)	758 (50%)	676 (51%)	1434 (50%)		
Age, years (mean, SD)	65.9 (7.4)	66.8 (7.2)	66.3 (7.4)	66.1 (7.7)	66.7 (7.9)	66.4 (7.8)		
Height, cm (mean, SD)	159.6 (7.9)	160.4 (8.1)	160 (8)	160.9 (7.9)	161.3 (8.2)	161.1 (8)		
Weight, kg (mean, SD)	66.1 (12.2)	65.8 (10.3)	66 (11.3)	64.8 (11)	65.8 (10.9)	65.3 (10.9)		
BMI, kg/m <sup>2</sup> (mean, SD)	25.9 (4.3)	25.5 (3.3)	25.7 (3.9)	25.0 (3.6)	25.2 (3.5)	25.1 (3.6)		
24-h sodium excretion, g/day (mean, SD)	4.4 (1.9)	4.2 (1.8)	4.3 (1.8)	3.8 (1.6)	4.0 (1.9)	3.9 (1.7)		
24-h potassium excretion, g/day (mean, SD)	1.4 (0.6)	1.4 (0.6)	1.4 (0.6)	2.1 (0.9)	1.4 (0.6)	1.8 (0.9)		

Table 1. Participant characteristics at baseline and follow-up, overall and for each randomized group

Intervention was use of reduced-sodium, added-potassium salt substitute. Control was continued use of usual salt.

not have been in any way affected by the reason for retraction, and was used because it was the best available external estimate.

Each of these methods (except for the estimating equations) was also used to estimate the change in 24-h potassium excretion between randomized groups from baseline to follow-up, since the salt substitute would be anticipated to also increase daily potassium intake. Findings for each method were compared by plotting point estimates of effect and 95% confidence intervals.

The 24-h urine samples were included only if: (1) less than 10% of the 24-h urine was self-reported as missing through spillage or uncollected voids; (ii) 24-h urine volume was between 500 ml and 6000 ml; (iii) 24-h creatinine excretion was between 4 mmol and 25 mmol for women, or 6 mmol and 30 mmol for men;<sup>21</sup> and (iv) 24-h urine collection duration was between 22 and 26 h (Figure 1). These are criteria for completeness of collection used in a previous study,<sup>22</sup> although they are unlikely to be as effective as the use of para-aminobenzoic acid (PABA),<sup>23–26</sup> which was not done in this project. In each case, a valid 24-h urine had to have an accompanying spot urine sample collected for the same participant at the same time point, for the data point to be included in the analyses.

For participants with baseline and follow-up urine samples, a paired analysis strategy was used to estimate the differences between baseline and follow-up excretion; and for the remainder, an unpaired methodology was used to control for baseline excretion levels. Overall differences were estimated by using a fixed effect meta-analytical approach to combine the data from the paired and unpaired analyses. To maximize power, the follow-up data were treated as a single time point regardless of whether measures were made at 6, 12 or 24 months. If participants were included in more than one follow-up survey, the last measurement was used. Analyses used SAS Enterprise version 7.1 and Stata version 13.1.

#### Results

There were 961, 298, 1778 and 4325 (total 7362) participants invited to participate in the baseline and 6-, 12- and 24-month follow-up surveys, respectively, with 4931 (67%) of invitations resulting in the collection of one or more urine samples. After exclusions (583 with only a spot urine sample or with only a 24-h urine sample, and 843 for other reasons; Figure 1) there were 3396 sets of 24-h urine collections and corresponding spot urine samples that contributed to the analyses. Of these 3396 sets of samples, 252 were paired samples collected from 126 individuals with measurements made at both baseline and follow-up, and 3144 were unpaired samples (406 collected at baseline and 2738 collected at follow-up). Characteristics of participants were broadly comparable across intervention and control groups and at baseline and follow-up (Table 1). There were two provinces (Liaoning and Shanxi) from which samples were only collected during the follow-up period.

# Estimated differences in sodium excretion between randomized groups

The mean difference in urinary sodium concentration in the intervention group compared with the control group



#### Figure 1. Flowchart.

was -14.3 mmol/l [95% confidence interval (CI): -23.5 to -5.1; P = 0.002] for the 3396 assays of 24-h urine samples, and -18.4 mmol/L (95% CI: -29.9 to -6.9; P = 0.002) for the 3396 assays of spot urine samples (Table 2). The 24-h urine volumes measured for the 3396 assays of 24-h urine samples were not different between randomized groups with a mean difference of 71 ml (95% CI: -50 to 192; P = 0.248).

The estimated difference in 24-h urinary sodium excretion between intervention and control groups was -0.35 g (95% CI: -0.68 to -0.02; P = 0.039) based upon the standard 24-h urine collections (Figure 2). The corresponding estimates obtained from the four published equations based upon spot urine samples were all smaller, and none identified a clear difference between groups (all P > 0.100), though point estimates of effects were all directionally similar (Figure 2). The estimate of difference using the 'individual volume spot' method was comparable in magnitude (-0.48 g), but no clear difference between groups was identified (95% CI: -0.98 to 0.02; P = 0.062). The estimate of difference based upon the 'standard volume spot', (-0.65 g, 95% CI: -1.06 to -0.25) was greater than that obtained from the 24-h urine collection and identified a difference between groups (P = 0.002). Estimates of the difference using the 'standard urine volume' approach with volumes 10% lesser or greater were -0.59 g (95% CI: -0.96 to -0.22) and -0.72 g (95% CI: -1.17 to -0.27), respectively (both P = 0.002).

# Estimated differences in potassium excretion between randomized groups

The difference in urinary potassium concentration in the intervention group compared with the control group was 11.7 mmol/l (95% CI: 8.6 to 14.8; P < 0.001) for the 3396 assays of the 24-h urine samples and 18.4 mmol/l (95% CI: 12.4 to 24.4; P < 0.001) for the 3396 spot urine samples. The estimated difference in 24-h urinary potassium

	Intervention			Control			Difference between	<i>P</i> -value
	Baseline	Follow-up	Change from baseline (95% CI)	Baseline	Follow-up	Change from baseline (95% CI)	intervention and control (95% CI)	
Spot urine elect	rolyte and creatinir	le concentrations (r	mmol/l)					
Potassium	43.9 (24.4)	67.2 (36.7)	22.1 (17.6 to 26.5)	42.6 (26.2)	47.9 (27.9)	5.4(1.7  to  9.1)	18.4 (12.4 to 24.4)	< 0.001
Sodium	130.6 (58.8)	123.6(58.4)	-7.8 (-15.5  to  -0.1)	118.5(55.6)	131.3(63.3)	12.1 (3.7  to  20.4)	-18.4 (-29.9 to -6.9)	0.002
Creatinine	10.1(6.5)	10.2(6.4)	0.3 (-0.6  to  1.1)	9.8 (6.8)	10.7~(6.7)	0.7 (-0.2  to  1.6)	-0.4 (-1.7  to  0.8)	0.519
24-h urine elect	rolyte and creatinir	ne concentrations (1	mmol/l)					
Potassium	23.6 (10.5)	40.1 (20.7)	15.8 (13.4 to 18.2)	22.5 (10.7)	26.6(14.3)	4.4 (2.7 to 6.2)	11.7 (8.6 to 14.8)	<0.001
Sodium	122.3 (46.6)	119.8(51.3)	-3.0(-9.1  to  3.1)	113.7 (47.7)	125.4 (51.9)	11.5 (4.8 to 18.3)	-14.3(-23.5  to  -5.1)	0.002
Creatinine	5.9(3.0)	6.5 (3.4)	0.6(0.2 to 1.0)	5.7 (2.7)	6.5(3.4)	0.7~(0.3 to 1.2)	-0.2 (-0.8  to  0.4)	0.612
24-h volume (n	(Ir							
Urine	1700.1 (713.4)	1512.3 (670.2)	-188.3 (-265.3  to  -111.2)	1757.2 (715.2)	1509.3~(689.7)	-244.6 (-334.8  to  -154.4)	71.0 (-50.0 to 192.0)	0.248
Values are exp	ressed as mean (SD) if	not otherwise specifie	ed.					

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excretion between intervention and control groups was 0.66 g (95% CI: 0.52 to 0.80; P < 0.001) based upon the standard 24-h urine collections. The estimates made using the 'individual volume spot' and the 'standard volume spot' methods all identified clear differences (all P < 0.001), though the point estimates of effect were all approximately 2-fold overestimates compared with those obtained from the 24-h urine collections (Figure 2).

#### Discussion

These data suggest significant potential for a greatly simplified approach to monitoring population sodium excretion, based upon the collection of spot urine samples rather than 24-h urine samples. Differences in population sodium and potassium excretion resulting from the administration of a reduced-sodium added-potassium salt substitute were clearly reflected in the mean concentrations of sodium and potassium in both spot and 24-h urine samples. Given no corresponding differences in mean urine volume between groups, it was possible to apply an externally derived estimate of mean population urine volume and detect a difference in urinary sodium and potassium excretion. Although the magnitude of the estimates in difference for mean population daily sodium and potassium excretion, obtained from the simple method based upon spot urine samples and a standard urine volume, varied in comparison with those obtained from the 24-h urine collections, the data provide initial proof of principle. Replication of the approach in other datasets will be required to confirm these findings and determine the extent to which methods based upon this approach might be biased or prone to random errors compared with methods based upon 24-h urine samples. If the findings are confirmed, then this would greatly simplify the measurement of population salt intake around the world and offer policy makers a practical, low-cost and easily implemented new way to monitor the effectiveness of intervention programmes.

The published equations that have been used to estimate the mean population level of sodium intake at a single time point were in this study ineffective for detecting differences in sodium intake between groups. The magnitudes of difference calculated were substantial underestimates compared with the results obtained from the 24-h urine. Previous cross-sectional analyses of daily excretion estimates based upon these equations showed that estimates based upon spot urine samples were able to provide reasonable approximations of sodium excretion at a given time point, but also that there was a tendency to underestimate excretion at high excretion levels and to overestimate excretion at low excretion levels.<sup>14</sup> Over- and understimation of this type would be expected to produce

Estimated difference of	of 24-hour sodium and potassium inta	ke from base	eline to follow	up (g)			Es	timated	differe	nce be	tween	interve	ntion and control (95% CI	P-value
	Intervention	Control												
Sodium														
Accepted standard measurement method	t i i i i i i i i i i i i i i i i i i i													
24-h urine sample														
Paired	-0.88	-0.39			_		-	_					-0.49 (-1.23, 0.26)	198
Unnaired	-0.55	-0.23				_	-						-0.31 (-0.68, 0.05)	095
Overall	-0.65	-0.25			-	•	-						-0.35 (-0.68, -0.02)	.039
Established estimating equations that use	e spot urine samples													
Tanaka														
Paired	-0.17	-0.05			_	_		_					-0.12 (-0.59, 0.36)	621
Unnaired	-0.10	-0.05					-						-0.05 (-0.26, 0.16)	653
Overall	-0.12	-0.05				_	-						0.06 (-0.26, 0.13)	541
Kawacaki	-0.12	-0.05											-0.00 (-0.20, 0.13)	.041
Deired	0.07	0.00				_	_		_				0 19 ( 0 06 0 60)	640
Paired	-0.27	-0.09					-						-0.18 (-0.96, 0.60)	.049
Unpaired	-0.17	-0.10				(i)	-						-0.07 (-0.42, 0.27)	.673
Overall	-0.19	-0.10					-	_					-0.09 (-0.41, 0.22)	.569
Intersalt														
Paired	-0.13	-0.05					-						-0.07 (-0.35, 0.20)	.587
Unpaired	-0.09	0.01											-0.10 (-0.31, 0.12)	.381
Overall	-0.11	-0.02				_							-0.09 (-0.26, 0.08)	.307
Intersalt with potassium														
Paired	-0.17	-0.05				_							-0 13 (-0 40 0 15)	363
Unpaired	-0.15	0.00				_	<b>⊢</b>						-0 15 (-0 37 0 07)	183
Overall	-0.16	-0.02				-	+						-0.14 (-0.31, 0.03)	.109
Simple calculations using spot urine sam	ple concentrations and urine different	volume estir	mates											
Individual volume spot	-0.87	-0.30		_	-		<u> </u>						-0.48 (-0.98, 0.02)	062
Standard volume spot	-0.28	0.43			-								-0.65 (-1.06 -0.25)	002
Standard volume spot - 10%	-0.25	0.30			_								-0.59 (-0.96 -0.22)	002
Standard volume spot + 10%	-0.31	0.47	2	1.20	•	_							-0.72 (-1.17, -0.27)	.002
	P-11-2-08			-	_		-	-	-	_	_	-		
			-1.2	9	6	3	0	.3	.6	.9	1.2	1.5	1.8	
Potassium														
Accepted standard measurement method	1	11010221												0.000 A.000
24-h urine sample	0.62	-0.02											0.66 (0.52, 0.80)	< 0.001
Simple calculations using spot urine sam	ple concentrations and urine different	volume estir	mates								_			
Individual volume spot	1.05	-0.14										C	1.29 (0.86, 1.72)	< 0.001
Standard volume spot	1.33	0.32									•	_	1.11 (0.75, 1.47)	< 0.001
Standard volume spot - 10%	1.20	0.30							-	-			1.00 (0.67, 1.33)	< 0.001
Standard volume spot + 10%	1.47	0.36									-		1.22 (0.82, 1.62)	< 0.001
						- 0		1		1	1	1	1	
			-1.2	- 9	- 6	- 3	0	.3	.6	.9	1.2	1.5	1.8	
						.0						1.0		

Figure 2. Estimated differences in 24-h urinary sodium and potassium excretion (g) between randomized groups based upon: (1) accepted standard of 24-h urine samples; (ii) published estimating equations that use spot urine samples; and (iii) simple calculations using spot urine samples and urine volume estimates. As 24-h urine collection is the most accepted method for measuring sodium intake and there were limited paired samples in our study, the overall estimate based on 24-h urine sample is considered the most plausible result.

Individual volume spot = sodium concentration in individual spot urine sample x volume of individual 24-h urine sample. Standard volume spot = sodium concentration in individual spot urine sample x 1.55 l. Standard volume spot -10% used a urine volume 10% less than 1.55 l (1.40 l). Standard volume spot +10% used a urine volume 10% greater than 1.55 l (1.71 l). Simple calculation of sodium and all estimates of potassium were only presented as overall values.

systematic underestimation of the true difference in excretion between two groups and may, in part at least, explain the smaller effects observed here.

The results of these analyses are aligned with the findings from a previous study that used data from two Australian States (New South Wales and Victoria) to estimate differences in sodium intake over time using spot and 24-h urine samples.<sup>27</sup> That study also showed that methods based upon spot urine samples were able to detect a difference in sodium excretion, although in that study the estimates of effect obtained were derived from the published equations, albeit only for the subset of analyses based on paired data. The published equations include multiple variables, and in unpaired analyses small differences in spot urine sodium concentrations may be masked by differences in covariates such as age, gender, body mass index and urine creatinine. In the paired analyses, by comparison, differences in such covariates would be removed or substantially reduced. The unpaired nature of the great majority of the data in the current report may explain why we were unable to detect differences using the published equations in contrast to the previous study. Paired samples were available for only 126 individuals in the present

study, and were too few to enable a robust assessment of effects in that subset.

The potential to detect differences in daily sodium (and potassium) excretion using simple calculations, based upon only spot urine electrolyte concentrations and an externally derived standard estimate of mean population urine volume, is important because it might preclude the need for assessment of urinary creatinine or direct measurement of 24-h urine volume. Urinary creatinine, unlike sodium and potassium concentrations, cannot easily be measured in the field, and 24-h urine volume measurement is onerous for participants and prone to error. Further, collecting paired data from the same individuals at multiple time points, as appears to be necessary for estimation of differences using the published equations, is difficult to achieve. Accordingly, a method based upon unpaired data with no requirement for direct measurement of 24-h urine volume would be preferable.

Statistical power to detect differences in mean population sodium excretion would require a greater sample size for methods based upon spot urine samples compared with 24-h urine samples. To detect a 0.35 g difference in sodium with 80% power and 95% confidence, using 24-h urine samples, would require a total sample size of 420 (210 per group) based upon paired samples and a sample size of 832 (416 per group) based upon unpaired samples, assuming a standard deviation of 1.8 g for sodium excretion. For methods based upon 'standard volume spot', corresponding power would be achieved with 624 samples for a study based upon paired samples and 1242 samples for a study based upon unpaired samples, assuming a standard deviation of 2.2 g for sodium excretion. In practice, because the collection of paired samples is likely to be impractical in most settings, unpaired analyses and larger samples will be required. Although the required sample size is larger, the much greater feasibility of spot urine sample collection may nonetheless make studies based upon unpaired spot urine samples more plausible.

Key strengths of this study are the large sample size and the standardized approach to the collection and analysis of both the spot and 24-h urine samples at multiple time points. The broad coherence of the findings for the spoturine based methods across both sodium and potassium provides reassurance that the main findings are unlikely to be attributable to chance. The generalizability of the results beyond rural China cannot be directly assessed, although the physiology underpinning sodium and potassium regulation is similar across diverse ethnic groups. Likewise the generalizability of the method for the assessment of sodium reduction strategies that use approaches other than salt substitution requires confirmation. The estimates obtained from the spot urine samples did not exactly match the estimates obtained from the 24-h urine samples, and further data are required to understand whether this reflects random errors or biases. If the latter, as seems likely for potassium at least, then adjustment to methods will need to be developed such that the magnitude of the difference derived from the spot urine-based methods more closely approximates that obtained from the 24-h urine samples. For example, the large difference between the concentrations of potassium in spot and 24-h urine specimens likely reflects the known diurnal variation of urinary potassium excretion, and this could be controlled for by specifying collection times and adjustments during analyses.<sup>28</sup> The simple methods described in this study depend upon there being no substantive difference in mean urine volume between the populations being compared, and there are reports indicating that a decrease in sodium intake can result in a decrease in 24-h urine volume.<sup>29</sup> Additional data describing the changes in mean population urine volume associated with changes in mean population salt intake, and confirmation that any changes in urine volume do not importantly bias estimates of change in salt intake, are needed. Also, since the external urine volume was a variance-free constant, the result did not incorporate any

allowance for variance or uncertainty in the volume estimate. Refinements that estimate mean population urine volume based upon characteristics such as age, sex and weight, or apply strata-specific estimates of urine volume, may enhance the methodology. Finally, these analyses provide a basis for the assessment of mean population sodium excretion but not for the sodium excretion level of an individual.

These data affirm earlier reports that spot urine-based methods may be effective not just for estimating mean population sodium excretion at a single time point, but also for measuring differences in mean sodium excretion between population samples.<sup>27</sup> The primary implication of this finding is that a much simpler and more practical method for monitoring the effectiveness of sodium reduction programmes may be feasible. Whereas there is a need for confirmation and refinement of the methods outlined here before they can be used at scale, these data provide a strong rationale for the further exploration of the approach.

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#### References

- Conlin PR. Eat your fruits and vegetables but hold the salt. *Circulation* 2007;116:1530–31.
- Law MR, Frost CD, Wald NJ. By how much does dietary salt reduction lower blood pressure? III—analysis of data from trials of salt reduction. *BMJ* 1991;302:819–24.
- Strazzullo P, D'Elia L, Kandala NB, Cappuccio FP. Salt intake, stroke, and cardiovascular disease: meta-analysis of prospective studies. *BMJ* 2009;339:b4567.
- WHO. Global Action Plan for the Prevention and Control of Noncommunicable Diseases 2013–2020. Geneva: World Health Organization (WHO), 2013.
- Filippini T, Violi F, D'Amico R, Vinceti M. The effect of potassium supplementation on blood pressure in hypertensive subjects: a systematic review and meta-analysis. *Int J Cardiol* 2017; 230:127–35.
- Simpson FO. Sodium intake, body sodium, and sodium excretion. *Lancet* 1988;2:25–29.
- 7. McLean RM. Measuring population sodium intake: a review of methods. *Nutrients* 2014;6:4651–62.
- Gibson RS. Principles of Nutritional Assessment. 2nd edn. Oxford, UK: Oxford University Press, 2005.
- Hawkes C, Webster J. National approaches to monitoring population salt intake: a trade-off between accuracy and practicality? *PLoS One* 2012;7:e46727.
- World Health Organization. European Regional Technical Consultation on Noncommunicable Disease Surveillance, Monitoring and Evaluation. Geneva: WHO, 2012.

- Huber DR, Blount BC, Mage DT, Letkiewicz FJ, Kumar A, Allen RH. Estimating perchlorate exposure from food and tap water based on US biomonitoring and occurrence data. J Expo Sci Environ Epidemiol 2011;21:395–407.
- Mage DT, Allen RH, Kodali A. Creatinine corrections for estimating children's and adult's pesticide intake doses in equilibrium with urinary pesticide and creatinine concentrations. *J Expo Sci Environ Epidemiol* 2008;18:360–68.
- Zimmermann MB, Andersson M. Assessment of iodine nutrition in populations: past, present, and future. *Nutr Rev* 2012;70: 553–70.
- Huang L, Crino M, Wu JH *et al*. Mean population salt intake estimated from 24-h urine samples and spot urine samples: a systematic review and meta-analysis. *Int J Epidemiol* 2016;45: 239–50.
- Neal B, Tian M, Li N *et al.* Rationale, design, and baseline characteristics of the Salt Substitute and Stroke Study (SSaSS)—a large-scale cluster randomized controlled trial. *Am Heart J* 2017;188:109–17.
- Brown IJ, Dyer AR, Chan Q *et al.* Estimating 24-h urinary sodium excretion from casual urinary sodium concentrations in Western populations: the INTERSALT study. *Am J Epidemiol* 2013;177:1180–92.
- 17. Kawasaki T, Itoh K, Uezono K, Sasaki H. A simple method for estimating 24 h urinary sodium and potassium excretion from second morning voiding urine specimen in adults. *Clin Exp Pharmacol Physiol* 1993;20:7–14.
- Tanaka T, Okamura T, Miura K *et al.* A simple method to estimate populational 24-h urinary sodium and potassium excretion using a casual urine specimen. *J Hum Hypertens* 2002;16:97–103.
- Chen X, Guo X, Ma J *et al.* Urinary sodium or potassium excretion and blood pressure in adults of Shandong province, China: preliminary results of the SMASH project. *J Am Soc Hypertens* 2015;9:754–62. http://onlinelibrary.wiley.com/o/cochrane/clcentral/articles/041/CN-01106041/frame.html (21 September 2018, date last accessed).

- 20. Retraction notice to "Urinary sodium or potassium excretion and blood pressure in adults of Shandong province, China: preliminary results of the SMASH project": [J Am Soc Hypertens 9 (2015) 754-762]. J Am Soc Hypertens 2016;10:472.
- Stolarz-Skrzypek K, Kuznetsova T, Thijs L *et al*. Fatal and nonfatal outcomes, incidence of hypertension, and blood pressure changes in relation to urinary sodium excretion. *JAMA* 2011; 305:1777–85.
- 22. Li N, Yan LL, Niu W *et al*. The effects of a community-based sodium reduction program in rural China—a cluster-randomized trial. *PLoS One* 2016;11:e0166620.
- John KA, Cogswell ME, Campbell NR *et al.* Accuracy and usefulness of select methods for assessing complete collection of 24h urine: a systematic review. *J Clin Hypertens* 2016;18:456–67.
- 24. De Keyzer W, Huybrechts I, Dekkers AL *et al.* Predicting urinary creatinine excretion and its usefulness to identify incomplete 24 h urine collections. *Br J Nutr* 2012;**108**:1118–25.
- 25. Subar AF, Kipnis V, Troiano RP *et al*. Using intake biomarkers to evaluate the extent of dietary misreporting in a large sample of adults: the OPEN study. *Am J Epidemiol* 2003;158:1–13.
- 26. Sadler K, Nicholson S, Steer T et al. National Diet and Nutrition Survey—Assessment of Dietary Sodium in Adults (Aged 19 to 64 Years) in England, 2011: A Survey Carried out on Behalf of the Department of Health. London: NatCen Social Research, University College London, MRC Human Nutrition Research, 2012.
- 27. Petersen KS, Wu JH, Webster J *et al.* Estimating mean change in population salt intake using spot urine samples. *Int J Epidemiol* 2017;**46**:1542–50.
- Hishida A, Honda N, Kumagai H, Sudo M, Nagase M. Diurnal variation of urinary potassium excretion in chronic renal insufficiency. *Miner Electrolyte Metab* 1982;7: 20-27.
- 29. He FJ, Markandu ND, Sagnella GA, MacGregor GA. Effect of salt intake on renal excretion of water in humans. *Hypertension* 2001;**38**:317–20.

### Appendix

Method	Urine sample	Formula to predict 24-h sodium intake (g)
Tanaka	Casual spot urine	$\begin{array}{l} 23 \div 1000 \times 21.98 \times \{\text{spot Na (mmol/l)/[spot creatinine (mg/dl) \times 10]} \times \\ [-2.04 \times \text{age (years)} + 14.89 \times \text{weight (kg)} + 16.14 \times \text{height (cm)} - \\ 2244.45]\}^{0.392} \end{array}$
Kawasaki	Second morning urine	
Male	-	$23 \div 1000 \times 16.3 \times \{\text{spot Na (mmol/l)/[spot Cr (mg/dl) x 10]} \times [-12.63 \times \text{age} (\text{years}) + 15.12 \times \text{weight (kg)} + 7.39 \times \text{height (cm)} - 79.9]\}^{0.5}$
Female		$23 \div 1000 \times 16.3 \times \{\text{spot Na (mmol/l)/[spot Cr (mg/dl) x 10]} \times [-4.72 \times \text{age} (\text{years}) + 8.58 \times \text{weight } (\text{kg}) + 5.09 \times \text{height } (\text{cm}) - 74.5]\}^{0.5}$
Intersalt with K	Casual spot urine	
Male		23 ÷ 1000 × {25.46 + [0.46 × spot Na (mmol/l)] – [2.75 × spot Cr (mmol/l)] – [0.13 × spot K (mmol/l)] + [4.10 × BMI (kg/m <sup>2</sup> )] + [0.26 × age (years)]}
Female		23 ÷ 1000 × {5.07 + [0.34 × spot Na (mmol/l)] – [2.16 × spot Cr (mmol/l)] – [0.09 × spot K (mmol/l)] + [2.39 × BMI (kg/m <sup>2</sup> )] + [2.35 × age (years)] – [0.03 × age <sup>2</sup> (years)]}
Intersalt without K	Casual spot urine	
Male		$23 \div 1000 \times \{23.51 + [0.45 \times \text{spot Na (mmol/l)}] - [3.09 \times \text{spot Cr (mmol/l)}] + [4.16 \times \text{BMI (kg/m}^2)] + [0.22 \times \text{age (years)}]\}$
Female		$23 \div 1000 \times \{3.74 + [0.33 \times \text{spot Na (mmol/l)}] - [2.44 \times \text{spot Cr (mmol/l)}] + [2.42 \times \text{BMI (kg/m^2)}] + [2.34 \times \text{age(years)} - [0.03 \times \text{age}^2(\text{years})]\}$

Table A1. Published pr	redictive equations that use	spot urine sodium conc	centration to estimate	24-h sodium intake
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