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Original article

The Perils of Sodium

Errors in estimating usual sodium intake by the Kawasaki formula alter its relationship with mortality: implications for public health†

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Abstract

Background: Several cohort studies with inaccurate estimates of sodium reported a J-shaped relationship with mortality. We compared various estimated sodium intakes with that measured by the gold-standard method of multiple non-consecutive 24-h urine collections and assessed their relationship with mortality.

Methods: We analysed the Trials of Hypertension Prevention follow-up data. Sodium intake was assessed in four ways: (i) average measured (gold standard): mean of three to seven 24-h urinary sodium measurements during the trial periods; (ii) average estimated: mean of three to seven estimated 24-h urinary sodium excretions from sodium concentration of 24-h urine using the Kawasaki formula; (iii) first measured: 24-h urinary sodium measured at the beginning of each trial; (iv) first estimated: 24-h urinary sodium estimated from sodium concentration of the first 24-h urine using the Kawasaki formula. We included 2974 individuals aged 30–54 years with pre-hypertension, not assigned to sodium intervention.

Results: During a median follow-up of 24 years, 272 deaths occurred. The average sodium intake measured by the gold-standard method was 3769 ± 1282 mg/d. The average estimated sodium over-estimated the intake by 1297 mg/d (95% confidence interval: 1267–1326). The average estimated value was systematically biased with over-estimation at lower levels and under-estimation at higher levels. The average measured

sodium showed a linear relationship with mortality. The average estimated sodium appeared to show a J-shaped relationship with mortality. The first measured and the first estimated sodium both flattened the relationship.

Conclusions: Accurately measured sodium intake showed a linear relationship with mortality. Inaccurately estimated sodium changed the relationship and could explain much of the paradoxical J-shaped findings reported in some cohort studies.

Key words: sodium intake, 24-h urinary sodium, Kawasaki formula, mortality, cohort study

Key Messages

- There is a strong linear relationship between sodium intake and blood pressure. Raised blood pressure is a leading cause of death worldwide.
- The relationship between sodium intake and mortality has recently been contested. The J-shaped relationship observed in some cohort studies may be partially caused by use of a biased or unreliable estimate of an individual's usual sodium intake, e.g. a single spot urine with the Kawasaki formula.
- Our study, for the first time, has compared the relationship between sodium intake and mortality, based on various methods to assess usual sodium intake, including estimates based on the Kawasaki formula (single and average of multiple days) and a single measured 24-h urinary sodium excretion, with the gold-standard method, i.e. the average of multiple non-consecutive measured 24-h urinary sodium excretions.
- The average measured sodium (gold standard) showed a direct linear relationship with mortality. The estimated sodium intakes changed this linear association. There appeared to be a J-shaped relationship between the average estimated sodium and mortality. The first estimated and the first measured sodium both flattened the relationship.
- Our findings indicate that inaccurate measurement of sodium intake could be an important contributor to the paradoxical J-shaped findings reported in some cohort studies. Epidemiological studies should not associate health outcomes with unreliable estimates of sodium intake.

Introduction

Raised blood pressure (BP) is a leading cause of death and disability, accounting for approximately 10.7 million deaths per year globally.[1,2] Dietary sodium intake is the major cause of raised BP.[3] Several lines of evidence including experimental studies in animals, epidemiological studies and randomized trials in humans have demonstrated a linear relationship between sodium intake and BP; within the range of 1200–4800 mg/d (3–12 g/d salt), the lower the sodium intake, the lower the BP.[3–9] Countries that have successfully reduced sodium intake have shown a reduction in population BP and cardiovascular disease (CVD) mortality.[10–12]

The current mean population sodium intake among adults in most countries is approximately 4000 mg/d (10 g/d salt).[13] The World Health Organisation (WHO) has recommended a 30% reduction in sodium intake by 2025, with an eventual target of <2000 mg/d (5 g/d salt) for all countries.[14] Several recent cohort studies have challenged the recommendations for a population-wide reduction in sodium intake, as these studies suggested that there was a J- or U-shaped relationship between sodium and outcome.[15–18] A pooled analysis of four cohort studies showed that, compared with sodium intake of 4000–5000 mg/d (10–12.5 g/d salt), lower and higher sodium intake both were associated with an increased risk of CVD and all-cause mortality.[17] Based on these results, a working group from the World Heart Federation, the European Society of Hypertension and the European Public Health Association have suggested that sodium intake should only be reduced in countries with sodium intake above 5000 mg/d (12.5 g/d salt).[19] In other words, for the majority of countries, there is no need to reduce sodium intake.[19] Not surprisingly, the J-shaped findings[15–18] have created controversy, as they cast doubt on the current public health policy on reducing sodium consumption to less than 2000 mg/d (5 g/d salt) by the WHO.[14]

These cohort studies, however, have methodological problems, such as reverse causality and measurement error, particularly the use of a single spot urine to estimate an

individual's usual sodium intake, which is inaccurate and unreliable.[20–22] Spot urine measures urinary sodium concentration that is determined not only by sodium intake, but also by various other factors such as fluid consumption, time of the day, duration and volume of the collection, individuals' posture,[20,23] as well as time and amount of sodium in the last meal consumed.[24] Furthermore, short-term sodium excretion is regulated by neural hormonal systems (sympathetic nervous system, angiotensin II and aldosterone) that are associated with cardiovascular outcomes.[25] Hence, the sodium concentration in spot urine samples, which do not reflect a steady state, may reflect cardiovascular risk relating to the control mechanisms for sodium excretion. Additionally, various formulae (e.g. Kawasaki) have been used to estimate 24-h urinary sodium excretion from spot urine and these formulae depend on the predicted creatinine excretion, which is calculated using a formula based on age, sex, height and weight.[26] Some of these variables are related to health outcomes and this could confound the relationship between sodium intake and outcomes. Twenty-four-hour urinary sodium is considered the gold standard for assessing sodium intake. Despite this, one single 24-h urinary sodium measurement does not reflect an individual's usual intake due to the large day-to-day variations in sodium consumption and excretion.[27,28] Multiple non-consecutive 24-h urine collections are needed to accurately assess an individual's usual sodium intake, which is essential for studies looking at the association between sodium and health outcomes.[29]

The Trials of Hypertension Prevention (TOHP) collected three to seven 24-h urine samples over 1.5–4 years.[30,31] A follow-up study at 10–15 years post trial showed a strong linear relationship between sodium intake and cardiovascular risk.[32] A more recent follow-up study at over 20 years post trial demonstrated a linear relationship between sodium intake and all-cause mortality.[33]

Given the importance of the public health recommendations on sodium intake and the confusion caused by some cohort studies,[15–18] we performed a secondary analysis of the recent TOHP follow-up data to study whether alternative estimates of sodium intake would alter the linear relationship between sodium and mortality, in particular, looking at the sodium intake estimated from urinary sodium concentration using the Kawasaki formula that has been widely used in the cohort studies that reported a J-shaped relationship between sodium intake and health outcomes.

Methods

The methods of the TOHP follow-up study were reported in detail elsewhere[33] and only methods relevant to the current analysis are described in brief here.

TOHP randomized trials and mortality follow-up

The original TOHP consisted of two phases. TOHP I was carried out between September 1987 and January 1990, and evaluated the effects of four supplement and three lifestyle interventions, including weight loss and sodium reduction, on BP in 2182 men and women aged 30–54 years with high normal BP.[31] TOHP II was carried out between December 1990 and March 1995, and evaluated the effects of sodium reduction and weight loss on BP using a 2×2 factorial design in 2382 men and women aged 30–54 years with pre-hypertension and a body mass index of 110–165% of desirable body weight.[30] During the randomized trial periods, five to seven 24-h urine collections were scheduled over 18 months in TOHP I and three to five 24-h urine collections during 3–4 years in TOHP II. Because of possibly short-term intervention-related changes in sodium consumption, individuals in the active sodium-reduction group (327 in TOHP I and 1191 in TOHP II) were excluded from the current analysis. Another eight participants in TOHP I and seven in TOHP II were excluded due to missing sodium excretion. A further 3 participants in TOHP I and 17 in TOHP II were excluded due to the occurrence of an incident CVD event or death during the period of exposure assessment (i.e. the trial period). Thus, 1844 participants in TOHP I and 1167 participants in TOHP II were included in the current analysis.

The trial periods constitute the baseline period for subsequent follow-up. Post-trial follow-up was conducted for mortality beginning at the end of each trial period. A search of the National Death Index was carried out, accruing death information through December 2013.

Outcome and exposure in the present analysis

The primary outcome is all-cause mortality at the longest follow-up. The secondary outcome is the accuracy of the different estimates of sodium intake.

The terms for the various exposures are described as follows:

i. 'Average measured sodium intake' (gold standard) was calculated as the mean of three to seven 24-h urinary sodium excretions measured during the trial periods. This is the best characterized measure of individuals' usual sodium intake and therefore considered as the gold-standard method.
ii. 'Average estimated sodium intake' was calculated as the mean of three to seven estimated 24-h urinary sodium excretions based on sodium concentration of three to seven 24-h urine collections during the trial periods using the Kawasaki formula.

iii. 'First measured sodium intake' was the first 24-h urinary sodium excretion measured at the beginning of each trial period.
iv. 'First estimated sodium intake' was the estimated 24-h urinary sodium based on sodium concentration of the first 24-h urine collection at the beginning of each trial period using the Kawasaki formula.

The Kawasaki formula[26] to estimate 24-h urinary sodium is:

$$\text{24h urinary sodium}\left[\frac{\text{mmol}}{\text{d}}\right] = 16.3 \times \sqrt{\frac{\text{sodium concentration from 24h urine}\left[\frac{mmol}{L}\right]}{\text{creatinine concentration from 24h urine}\left[\frac{mg}{dL}\right] \times 10} \times \text{Predicted urinary creatinine}\left[\frac{mg}{d}\right]},$$

where the predicted urinary creatinine for females is as follows: creatinine (mg/d)=[–4.72×age (years)]+[8.58×weight (kg)]+[5.09×height (cm)]–74.5; and for males is as follows: creatinine (mg/d)=[–12.63×age (years)]+[15.12×weight (kg)]+[7.39×height (cm)]–79.9. Sodium (mg/d) was calculated as sodium (mmol/d) multiplied by 23.

Statistical analysis

Paired *t*-tests were carried out to compare the measured with the estimated 24-h urinary sodium for both the average and the first values. Among the 3011 participants included in our study, 355 did not collect the first 24-h urine (i.e. at baseline of the TOHP trials) and they were excluded from the analysis when using the first measured and the first estimated 24-h urinary sodium. In another participant, the average estimated 24-h urinary sodium could not be calculated due to missing urinary creatinine concentration. This participant was therefore excluded from the paired comparison of average measured with average estimated 24-h urinary sodium. A Bland-Altman plot was used to compare agreement of the estimated sodium intake with the measured value. The measured sodium intake was grouped into categories of <2300, 2300 to <3600, 3600 to <4800 and ≥4800 mg/d, where 2300 mg/d represents the currently recommended level for adults in the UK[34] and USA[35] and 3600 mg/d represents the median sodium intake in the US population aged 31–50 years.[36] For the estimated sodium intake, due to over-estimation by the Kawasaki formula, only one participant had an average estimated sodium intake of <2300 mg/d. Therefore, the estimated sodium intake was grouped into categories of <3600, 3600 to <4800, 4800 to <6000 and ≥6000 mg/d. Baseline characteristics were reported as percentages or means and were tested for trend over sodium categories using chi-square statistics or regression analysis.

Cox regression analysis was performed to estimate the association between sodium intake and mortality. Separate analysis was carried out for sodium intake as a continuous variable and in categories. The Cox regression models were stratified by study phase and all models were adjusted for clinic, age, sex, race/ethnicity, other treatment assignments, education, baseline weight, alcohol use and amount, smoking, exercise, potassium excretion and family history of CVD. Differences in linear slopes for various sodium estimates were tested using standard errors based on 1000 bootstrap samples. Penalized splines with 4 degrees of freedom were fit to examine linearity of effect in models adjusting for the same factors. Both these and the Bland-Altman plots were fitted using R. All other analyses were conducted using SAS version 9.3 (SAS, Cary, NC).

Results

Sodium intake

There were 1844 participants in TOHP I and 1167 in TOHP II who had not been randomized to a sodium-reduction intervention, with 37 participating in both phases, and therefore 2974 unique participants were in follow-up. Of these, 68% were men, 16% were African American and the mean age was 43 years. The mean number of 24-h urine collections during the trial periods was 4.4 in TOHP I and 3.5 in TOHP II.

The distributions of sodium intake (i.e. 24-h urinary sodium) estimated by the four different methods are shown in Figure 1. The overall mean (±SD) measured 24-h urinary sodium excretion during the trial periods was 3769 ± 1282 mg/d (9.42 ± 3.21 g/d salt). The mean estimated 24-h urinary sodium was 5066 ± 843 mg/d (12.67 ± 2.11 g/d salt). Paired comparison showed that the estimated 24-h urinary sodium over-estimated the sodium level by 1297 mg/d [95% confidence interval (CI): 1267–1326] [salt: 3.24 g/d (95% CI: 3.17–3.32)]. Similarly, the first estimated 24-h urinary sodium excretion over-estimated the measured value by 1088 mg/d (95% CI: 1042–1134) [salt: 2.72 g/d (95% CI: 2.61–2.84)] (Table 1). Subgroup analysis showed that, compared with the measured sodium levels, the estimated values were consistently higher in men and women in TOHP I and TOHP II. The degree of over-estimation was larger in TOHP I compared with TOHP II (Table 1).

The Bland-Altman plot showed poor agreement between the estimated and measured 24-h urinary sodium

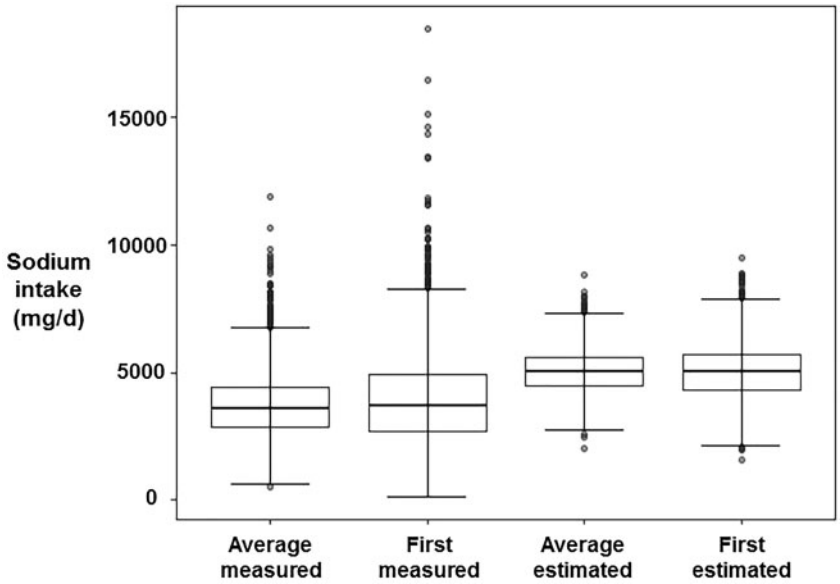

Figure 1. Box plots showing the distributions of sodium intake estimated by various methods. The lower and upper hinges correspond to the first and third quartiles (i.e. the 25th and 75th percentiles, respectively). The upper whisker extends from the hinge to the highest value no further than 1.5 × IQR from the hinge (where IQR is the inter-quartile range, or distance between the first and third quartiles). The lower whisker extends from the hinge to the lowest value no further than 1.5 × IQR of the hinge. Data beyond the end of the whiskers are 'outlying' points and are plotted individually. $P < 0.001$, $N = 3010$ for comparison of average measured vs average estimated. $P < 0.001$, $N = 2656$ for comparison of first measured vs first estimated.

excretion. The estimated sodium was systematically biased with over-estimation at lower levels and under-estimation at higher levels (Figures 2 and 3).

The mean of the first measured 24-h urinary sodium was 3940 ± 1813 mg/d (9.85 ± 4.53 g/d salt) in all participants. The distribution of the first measured 24-h urinary sodium was similar to that of the average measured value, but with a 40% increase in the standard deviation (Figure 1 and Table 1).

Association with mortality

Baseline characteristics by categories of sodium intake with various estimates are shown in Supplementary Tables 1–4, available as Supplementary data at *IJE* online, including age, sex, ethnic group, percentage of college education, smokers, alcohol drinkers and exercise of at least once per week, body weight, BP and average measured and estimated 24-h urinary sodium, as well as first measured and estimated 24-h urinary sodium. During the post-trial follow-up, 272 deaths occurred, 189 among TOHP I participants and 83 among TOHP II participants (Table 2). Median follow-up time following the trial periods among survivors was 23.9 years for TOHP I and 18.8 years for TOHP II.

Figure 4 shows the spline plots of different estimates of sodium intake and mortality with the rug marks on the *x*-axis indicating distribution of sodium. There was a significant linear association between the average measured sodium and mortality with a *P*-value for linearity of 0.047 and of 0.87 for deviation from linearity. Within the range of 1200–9000 mg/d (3.00–22.50 g/d salt), the lower the sodium intake, the lower the risk (Figure 4a), although the data were sparse and 95% CIs were wide at both the lowest and highest levels. Compared with sodium intake of 3600 to <4800 mg/d (9.00–12.00 g/d salt), the hazard ratios for sodium levels of 2300 to <3600 (5.75–9.00 g/d salt) and <2300 mg/d (5.75 g/d salt) were 0.95 and 0.75, respectively, but the trend across categories did not reach statistical significance (Table 2). When sodium was entered as a continuous variable, a 1000-mg/d higher sodium level was associated with a 12% increase in mortality with borderline statistical significance ($P = 0.052$).

Figure 4b shows the association of the average estimated 24-h urinary sodium with mortality. Due to the over-estimation of sodium at lower levels and under-estimation at higher levels, the distribution of sodium was shifted towards the mean value. The spline plot suggested a J-shaped relationship between the average estimated 24-h urinary sodium and mortality. Compared with estimated sodium levels of 3600 to <4800 mg/d (9.00–12.00 g/d salt), both lower and higher sodium levels appeared to be associated with an increased risk (Figure 4b and Table 3). Although the trend analysis was statistically significant for linearity, data were sparse at lower estimated sodium levels and the test for deviation from linearity was not significant.

Table 1. Comparison of measured 24-h urinary sodium with estimated 24-h urinary sodium based on sodium concentration using the Kawasaki formula

	Average 24-h urinary sodium (mg)					First 24-h urinary sodium (mg)				
	N	Measured	Estimated	Paired difference		*N*	Measured	Estimated	Paired difference	
		Mean ± SD	Mean ± SD	Mean (95% CI)	*P*-value		Mean ± SD	Mean ± SD	Mean (95% CI)	*P*-value
TOHP I										
Men	1290	3855 ± 1279	5305 ± 769	1450 (1403–1498)	<0.001	1071	4038 ± 1882	5251 ± 1042	1213 (1135–1291)	<0.001
Women	553	2998 ± 1043	4405 ± 657	1407 (1348–1466)	<0.001	451	2979 ± 1456	4333 ± 916	1354 (1263–1446)	<0.001
Combined	1843	3598 ± 1274	5035 ± 844	1437 (1400–1475)	<0.001	1522	3724 ± 1831	4979 ± 1090	1255 (1194–1316)	<0.001
TOHP II										
Men	769	4378 ± 1233	5418 ± 757	1040 (980–1099)	<0.001	750	4547 ± 1791	5383 ± 993	836 (750–923)	<0.001
Women	398	3386 ± 987	4527 ± 655	1141 (1073–1209)	<0.001	384	3609 ± 1481	4526 ± 921	917 (818–1017)	<0.001
Combined	1167	4040 ± 1247	5114 ± 838	1074 (1029–1120)	<0.001	1134	4229 ± 1749	5093 ± 1050	864 (797–930)	<0.001
All	3010	3769 ± 1282	5066 ± 843	1297 (1267–1326)	<0.001	2656	3940 ± 1813	5028 ± 1074	1088 (1042–1134)	<0.001

The first measured 24-h urinary sodium showed no association with mortality, although the spline plot suggested an inverse association at a sodium level of >8000 mg/d (20.00 g/d salt) where the sample size was very small (Figure 4c). The first estimated 24-h urinary sodium appeared to be associated with mortality only at sodium levels of ≥6500 mg/d (16.25 g/d salt) and, at sodium levels of <6500 mg/d, the spline curve was flat (Figure 4d). The trend analysis showed no significant association of the first measured or the first estimated 24-h urinary sodium with mortality (Tables 2 and 3).

Paired comparisons of the linear slopes of sodium to mortality showed that there was a significant difference in slopes between the average measured sodium (gold standard) and the average estimated value ($P = 0.043$), and a significant difference between the average measured sodium and the first measured value ($P = 0.035$). However, there was no significant difference in slopes between the average measured sodium and the first estimated value ($P = 0.68$) or between the first measured sodium and the first estimated value ($P = 0.0674$).

Discussion

Our study produced several important findings. First, the 24-h urinary sodium estimated from sodium concentration using the Kawasaki formula showed poor agreement with the measured value, with higher means and reduced variability. The estimation was systematically biased with over-estimation at lower levels and under-estimation at higher levels. Second, the estimated sodium intake altered the direct linear relationship between sodium and mortality observed in the TOHP follow-up study where the gold-standard method (i.e. multiple non-consecutive 24-h urine collections) was used. The average estimated sodium erroneously suggested a J-shaped relationship with mortality and the single estimated sodium flattened the relationship between sodium and mortality. Third, the distribution of a single measured 24-h urinary sodium was close to that of the average of three to seven 24-h urinary sodium excretions for mean sodium intake at the population level, though it had wider variability. Likely because of the increased measurement error, the use of a single measured 24-h urinary sodium excretion to estimate sodium intake flattened the linear association between sodium and mortality observed in the TOHP follow-up study.

For association studies on sodium and health outcome, accurate and reliable measurement of individuals' usual sodium intake matters.[29] Over the past five decades, there have been numerous attempts to try to simplify the measurements of individuals' daily sodium intake. However, carefully controlled studies showed that, to get a reasonable

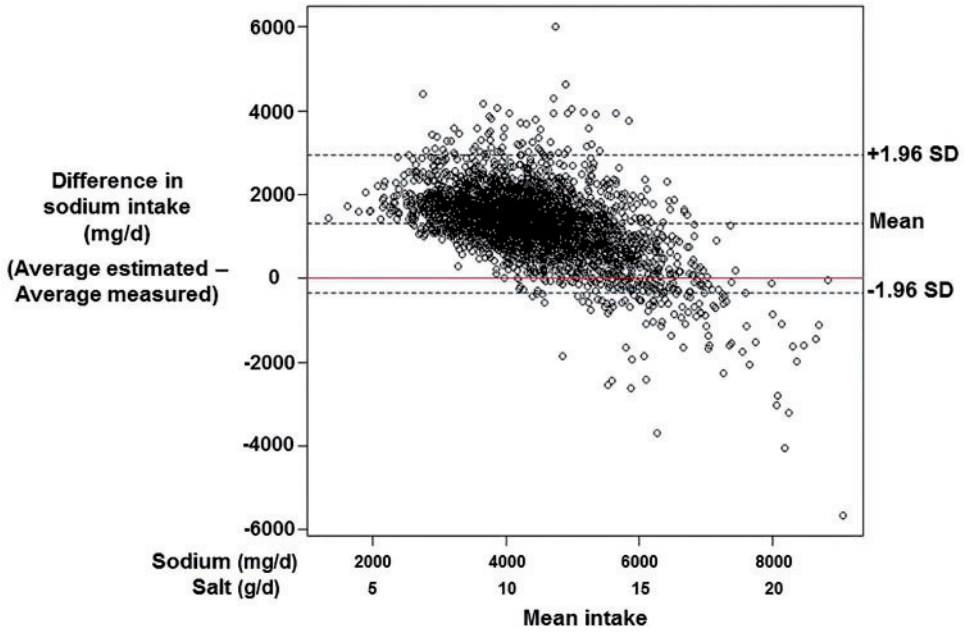

Figure 2. Bland-Altman plot comparing average estimated with average measured 24-h urinary sodium excretion during the trial periods.

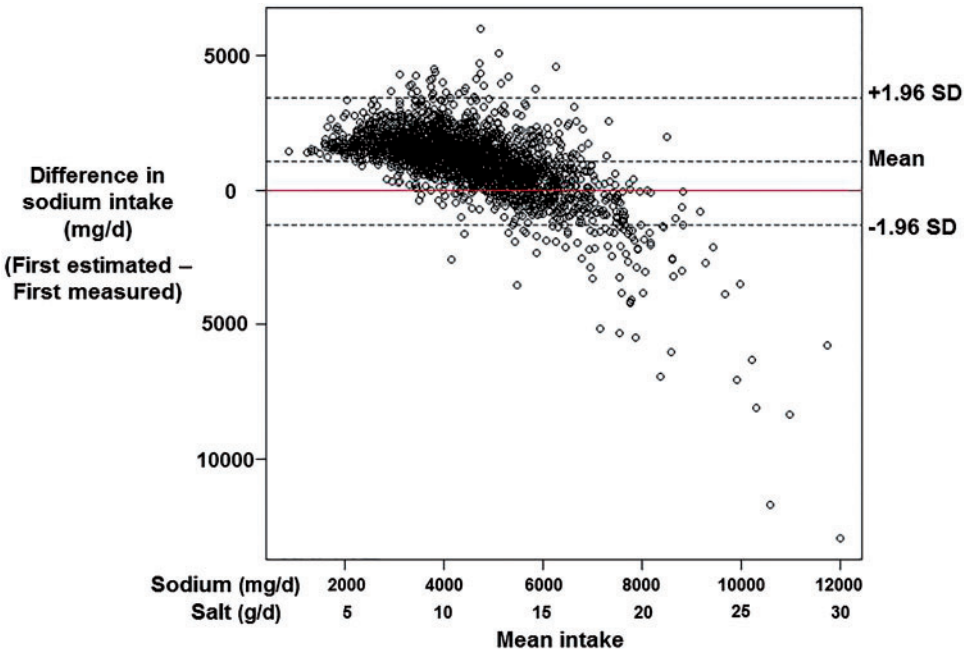

Figure 3. Bland-Altman plot comparing first estimated with first measured 24-h urinary sodium excretion at the beginning of each trial.

estimate in one individual, at least three 24-h urine collections would need to be made, due to the large day-to-day variations in sodium consumption and excretion.[27,28,37,38] The TOHP study collected up to seven 24-h urine samples and therefore provided the most accurate assessment of usual sodium intake in relation to outcomes to date. The TOHP follow-up study demonstrated a direct linear relationship between sodium intake and mortality down to a sodium level of 1200 mg/d (3 g/d salt), which is the long-term target for population sodium intake in the UK recommended by NICE (National Institute for Health and Care Excellence).[39] These findings are consistent with the evidence that there is a strong direct linear relationship between sodium intake and BP within the range of 1200–4800 mg/d[4,5,8,9] and a strong continuous graded relationship between BP and CVD, as well as mortality.[40,41]

Table 2. Total mortality in the TOHP cohorts post-trial follow-up by categories of measured 24-h urinary sodium excretion among individuals not in a sodium-reduction intervention

	Measured sodium excretion (mg/24 h)				*P* trend*	Hazard ratios per 1000 mg/24 h	*P*-value
	<2300	2300 to<3600	3600 to<4800	≥4800			
Average measured 24-h urinary sodium							
Deaths/total (%)							
TOHP I	22/246 (8.9)	73/775 (9.4)	63/566 (11.1)	31/257 (12.1)			
TOHP II	1/66 (1.5)	32/407 (7.9)	30/413 (7.3)	20/281 (7.1)			
Hazard ratios (95% CI)	0.75 (0.45–1.26)	0.95 (0.70–1.29)	1.00 (Reference)	1.07 (0.75–1.54)	0.304	1.12 (1.00–1.26)	0.052
First measured 24-h urinary sodium							
Deaths/total (%)							
TOHP I	33/319 (10.3)	47/475 (9.9)	41/380 (10.8)	38/351 (10.8)			
TOHP II	11/121 (9.1)	24/336 (7.1)	16/313 (5.1)	30/365 (8.2)			
Hazard ratios (95% CI)	1.18 (0.77–1.82)	1.13 (0.78–1.62)	1.00 (Reference)	1.23 (0.85–1.77)	0.921	1.00 (0.92–1.09)	0.946

From Cox proportional hazards regression models stratified by trial phase and adjusted for age, sex, race/ethnicity, clinic, treatment assignment, education status, baseline weight, alcohol use, smoking, exercise, 24-h urinary potassium excretion and family history of cardiovascular disease.

CI, confidence interval; TOHP, Trials of Hypertension Prevention. **P* for trend was tested by entering into the model the four categories of 24-h urinary sodium as a continuous variable.

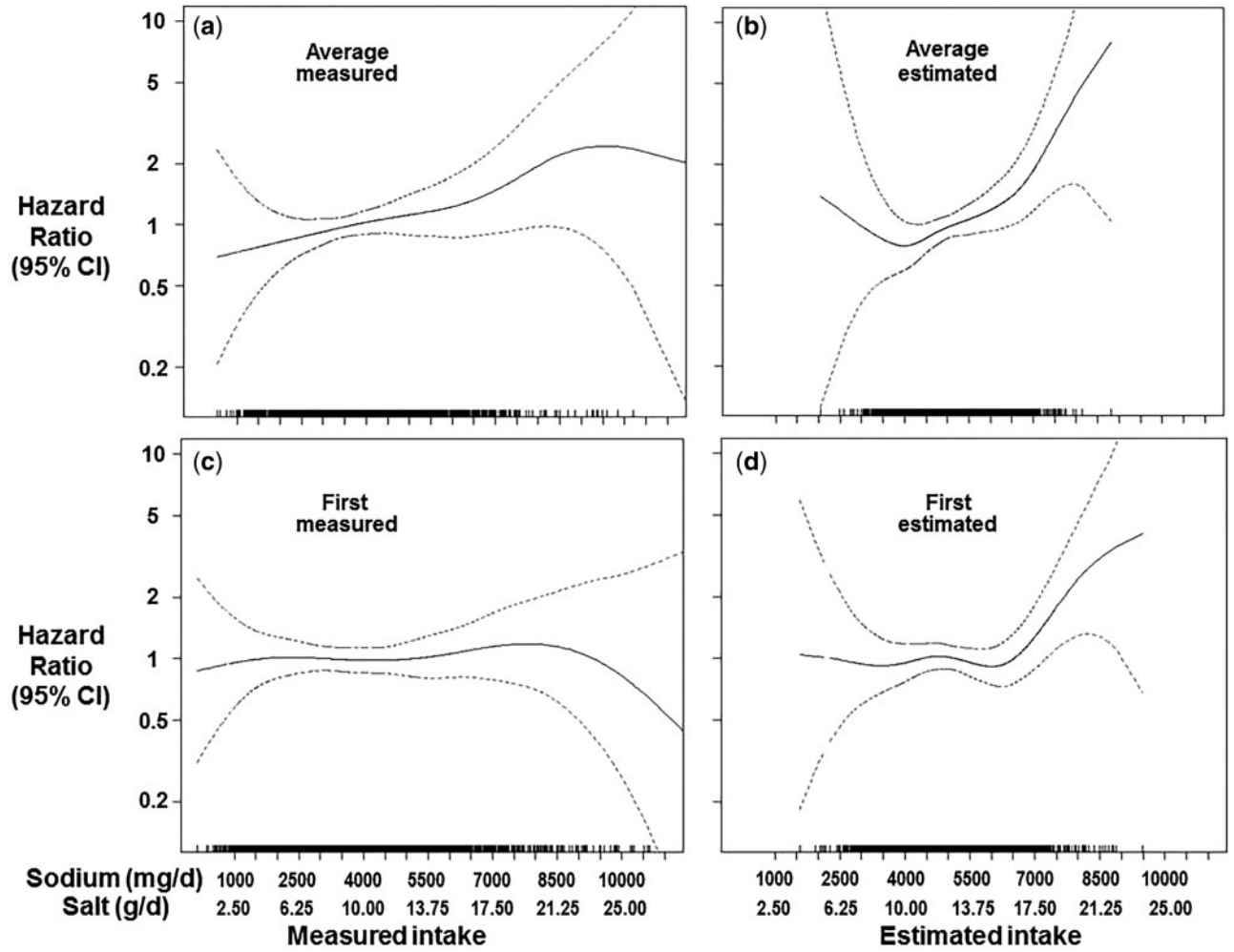

Figure 4. Spline plots for the association between different estimates of 24-h urinary sodium excretion and all-cause mortality adjusted for age, sex, race/ethnicity, clinic, treatment assignment, education status, baseline weight, alcohol use, smoking, exercise, 24-h urinary potassium excretion and family history of cardiovascular disease. Rug plot indicates distribution of sodium excretion. (a) *P* (Linear) = 0.047; *P* (Nonlinear) = 0.87. (b) *P* (Linear) = 0.006; *P* (Nonlinear) = 0.26. (c) *P* (Linear) = 0.82; *P* (Nonlinear) = 0.71. (d) *P* (Linear) = 0.15; *P* (Nonlinear) = 0.088.

Table 3. Total mortality in the TOHP cohorts post-trial follow-up by categories of estimated 24-h urinary sodium excretion using the Kawasaki formula among individuals not in a sodium-reduction intervention

	Estimated sodium excretion (mg/24-h)				*P* trend*	Hazard ratios per 1000 mg/24 h	*P*-value
	<3600	3600 to <4800	4800 to <6000	≥6000			
Average estimated 24-h urinary sodium							
Deaths/total (%)							
TOHP I	7/62 (11.3)	62/686 (9.0)	92/862 (10.7)	28/233 (12.0)			
TOHP II	1/27 (3.7)	23/414 (5.6)	40/556 (7.2)	19/170 (11.2)			
Hazard ratios (95% CI)	1.26 (0.60–2.65)	1.00 (Reference)	1.40 (1.01–1.94)	1.84 (1.17–2.90)	0.017	1.29 (1.06–1.57)	0.011
First estimated 24-h urinary sodium							
Deaths/total (%)							
TOHP I	12/154 (7.8)	54/524 (10.3)	73/659 (11.1)	26/269 (9.7)			
TOHP II	7/75 (9.3)	23/379 (6.1)	31/471 (6.6)	20/209 (9.6)			
Hazard ratios (95% CI)	0.89 (0.53–1.49)	1 (Reference)	1.03 (0.75–1.41)	1.12 (0.74–1.71)	0.480	1.09 (0.94–1.26)	0.243

From Cox proportional hazards regression models stratified by trial phase and adjusted for age, sex, race/ethnicity, clinic, treatment assignment, education status, baseline weight, alcohol use, smoking, exercise, 24-h urinary potassium excretion and family history of cardiovascular disease.

CI, confidence interval; TOHP, Trials of Hypertension Prevention. **P* for trend was tested by entering into the model the four categories of 24-h urinary sodium as a continuous variable.

On the contrary, several cohort studies have reported a J-shaped relationship, i.e. compared with sodium intake of 4000–5000 mg/d (10–12.5 g/d salt), both lower and higher intakes were associated with an increased risk of CVD and all-cause mortality.[15–17] These studies, however, have a number of methodological problems, one of which is error in the assessment of sodium intake, e.g. estimated sodium intake from spot urine using the Kawasaki formula. Studies that have directly compared sodium intake estimated from spot urine with that measured in 24-h urine have shown that estimates from spot urine are unreliable, not reproducible and systematically biased.[20–22] This consistent evidence that spot urine samples are not a valid method of estimating 24-h urine sodium is the basis for recommendations not to use spot urine to assess an individual's sodium intake.[42]

In the TOHP studies, spot urine was not collected. However, one of the assumptions in using spot urine to estimate sodium intake is that the ratio of spot urinary sodium concentration to creatinine concentration is about equivalent to the ratio of 24-h urinary sodium concentration to creatinine concentration.[26] Our analysis 'mimicked' estimates from spot urine in so far as using sodium concentration from 24-h urine to calculate the estimated sodium intake. As such, our estimates are likely to be more closely correlated with total 24-h urinary sodium excretion compared with any of the estimates from spot urine (e.g. casual, morning fasting void). Despite this, our results show that the estimated sodium intake using the Kawasaki formula is inaccurate and systematically biased compared with the measured sodium intake. Importantly, replacing the accurately measured average sodium with the estimated value appears to change the relationship between sodium and mortality from linear to J-shaped. This suggests that errors in assessing sodium intake with spot urine using the Kawasaki formula could be an important contributor to the J-shaped relationship observed in some of the cohort studies.[15–17]

In our analysis, we used the Kawasaki formula because it is the most commonly used in cohort studies and showed a J-shaped relationship between the estimated sodium intake and the risk of CVD and all-cause mortality.[15–17] Several other formulae have also been reported in the literature, although studies have shown that none is unbiased in estimating an individual's usual sodium intake, which is essential for association studies.[43,44]

Our study also showed that the degree of bias for the estimated sodium was larger in TOHP I compared with TOHP II. This is likely to be partly due to the difference between TOHP I and TOHP II participants in body weight that was used in the Kawasaki formula, as other variables in the formula, such as age and sex, were comparable between TOHP I and TOHP II.[32,33] Another contributor could be the difference in sodium intake between TOHP I and TOHP II, as the estimated values over-estimated sodium intake at lower levels and under-estimated intake at higher levels. Due to the limited sample size, we did not have the statistical power to separate analyses for these two studies in relation to mortality.

Twenty-four-hour urinary sodium excretion is considered the gold standard for assessing sodium intake. However, owing to the large day-to-day variations in sodium intake, one single measure does not reflect an individual's usual intake. Additionally, the variation of sodium

consumption within individuals is as much as that within the population.[27,38] It is therefore not surprising that our study found that a single measured 24-h urinary sodium flattened the linear relationship between sodium and mortality observed in the TOHP follow-up study.

Reverse causality is another potentially important contributor to the higher risk of CVD and mortality with lower sodium intake observed in some cohort studies, as many of them included people with vascular disease or at high risk of CVD (e.g. with chronic kidney disease or diabetes),[17] and therefore represented a population who were ill and were likely to eat less food and consume less sodium, or they were advised to reduce sodium intake and had made an effort to do so. As such, the inverse relationship between sodium and outcome could be the consequence of the underlying disease rather than the lower sodium intake. In contrast, the TOHP used careful screening and exclusion of participants with pre-existing CVD or other diseases that have the potential to affect both diet and mortality. Therefore, reverse causality is unlikely to have been an issue in the TOHP follow-up study.

The strength of our analysis includes (i) the TOHP collected multiple non-consecutive 24-h urine samples using standardized methods as part of a clinical trial,[32,33] which would have reduced the potential for systematic bias and random error in sodium assessment; (ii) TOHP cohorts were restricted to healthy, free-living, pre-hypertensive individuals,[32,33] which would have minimized the potential for reverse causality.

The limitations of our analysis include: (i) spot urine was not collected in the TOHP and we used sodium concentration from 24-h urine to calculate the estimated sodium intake; our estimated sodium was therefore more closely related to the measured 24-h urinary sodium excretion than spot urine, so it is possible that our results may have been conservative, under-estimating the random and systematic measurement error with spot urine; (2) the sample size, especially the number of endpoints at the lower sodium levels, was small and therefore our analysis had limited statistical power to test the relationship between sodium intake and outcome, which is particularly true for the estimated sodium intake due to the over-estimation at lower levels; this could explain the statistically non-significant result for deviation from linearity, but instead significant for linear, when the curve appeared J-shaped for the relationship between the average estimated sodium intake and mortality; (iii) the data for CVD events were not available in the latest TOHP follow-up at 19–24 years, but were recorded in the previous follow-up at 10–15 years; we did perform an analysis on CVD events, but there was only one CVD event in the category with the average estimated sodium intake of less than 3600 mg/d (9.0 g/d salt); such a small sample size would not allow any conclusion to be drawn. We therefore did not report the results in this paper; nevertheless, as CVD is the major cause of deaths in the USA, accounting for about one in three deaths,[45] it would have been the major component of total mortality in our study; (iv) similarly to almost all other cohort studies, no measurement of sodium intake was made during follow-up, so baseline exposure may not capture changes in intake over time; a recent study has suggested that the use of long-term multi-year 24-h urine collections significantly strengthened the association between sodium intake and CVD and renal risk.[46]

In conclusion, our analysis, by comparing various estimates of sodium intake with the gold-standard method of careful collections of multiple non-consecutive 24-h urine samples over a period of 18 months to 4 years in the TOHP studies, demonstrates that inaccurate estimates of sodium intake changed its relationship with mortality and could explain much of the paradoxical J-shaped findings reported in some cohort studies.[15–17] It is therefore vital to accurately assess individuals' usual sodium intake for association studies.

The finding of a direct linear relationship between the accurately measured sodium and mortality provides further support to the strong evidence for population-wide reduction in sodium intake.[3] Paradoxical results from methodologically flawed studies should not be used to derail critical public health policy, nor divert action. Gradual stepwise sodium reduction as recommended by the WHO remains an achievable, affordable, effective and important strategy to prevent CVD and premature deaths worldwide.

Supplementary data

Supplementary data are available at *IJE* online.

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