Clinical Nutrition xxx (xxxx) xxx

Contents lists available at ScienceDirect

Clinical Nutrition

journal homepage: http://www.elsevier.com/locate/clnu

Original article

Impact of urinary sodium on cardiovascular disease and risk factors: A 2 sample Mendelian randomization study

Shiu Lun Au Yeung [a, *], Catherine Mary Schooling [a, b]

[a] School of Public Health, Li Ka Shing Faculty of Medicine, The University of Hong Kong, Hong Kong, China
[b] City University of New York, Graduate School of Public Health and Health Policy, New York, USA

ARTICLE INFO

Article history:
Received 6 April 2020
Accepted 11 September 2020

Keywords:
Atrial fibrillation
Heart failure
Stroke
Sodium
Mendelian randomization

SUMMARY

Background: Although sodium increases the risk of coronary artery disease and hypertension, whether sodium also impacts other cardiovascular disease (CVD) and its risk factors is less clear. We examined the causal role of urinary sodium in these CVDs and risk factors using Mendelian randomization.

Methods: We identified strong, independent single nucleotide polymorphisms (SNPs) of urinary sodium from the most up to date genome wide association studies (GWAS) (n = 446,237) and applied them to GWAS of stroke and its subtypes (40,585 cases and 406,111 non-cases), atrial fibrillation (60,620 cases and 970,216 non-cases) and heart failure (47,309 cases and 930,014 non-case). We assessed the impact of sodium on these diseases and associated risk factors using inverse variance weighting. Sensitivity analyses included weighted median, contamination mixture method, MR-PRESSO, and multivariable Mendelian randomization.

Results: Higher log transformed urinary sodium was associated with higher risk of stroke (odds ratio (OR) 1.45, 95% confidence interval (CI) 1.01 to 2.08), ischemic stroke (OR 1.60 95% CI 1.12 to 2.30), heart failure (OR 1.77 95% CI 1.19 to 2.62), and type 2 diabetes (OR 4.17 95% CI 1.53 to 11.35). Sensitivity analyses produced directionally similar estimates.

Conclusion: Higher sodium likely increases stroke, heart failure and type 2 diabetes risk. Our study further supports public health policies to minimize population sodium intake, so as to reduce the associated disease burden.

1. Introduction

High intake of sodium is a major public health burden resulting in 3 million deaths and 70 million disability adjusted life years according to the Global Burden of Disease Study [1]. Whilst higher sodium intake is associated with increased risk of hypertension [2] and cardiovascular disease (including stroke, atrial fibrillation and heart failure) in observational studies in the general population and patient subgroups [3–7], some studies have suggested salt may not be an important determinant of cardiovascular disease [8]. However, given the evidence concerning the effects of salt on cardiovascular disease is mostly observational [9], confounding, reverse causation, or selection bias could drive these positive findings [3]. Experimental evidence suggests a reduction in blood pressure with reduced salt intake [10], but not with reduced lipids [11]. An experimental study also suggested a reduction in cardiovascular mortality when conventional salt was replaced by potassium-enriched salt, however it is difficult to assess whether the protective effect was due to the reduction of sodium, increase in potassium, or both [12].

In the absence of evidence from randomized controlled trials, the use of Mendelian randomization may help validate the findings from previous observational studies [13]. Mendelian randomization makes use of the random allocation of genetic variants related to exposure, and hence is less susceptible to confounding than observational studies. Recent Mendelian randomization studies confirmed the detrimental impact of urinary sodium, assessed from spot urine samples, on blood pressure, adiposity, and coronary artery disease [14], as well as the impact of related urinary biomarkers, such as urinary sodium-potassium ratio and urinary albumin-creatinine ratio, on blood pressure and diabetes [15]. However, it remains unclear whether higher urinary sodium also affects other

* Corresponding author. School of Public Health, Li Ka Shing Faculty of Medicine, The University of Hong Kong, 7 Sassoon Road, Pokfulam, Hong Kong, China. Fax: +852 3520 1945.
E-mail address: ayslryan@hku.hk (S.L. Au Yeung).

https://doi.org/10.1016/j.clnu.2020.09.018
0261-5614/

important cardiovascular outcomes, such as stroke, atrial fibrillation and heart failure, given the lack of relevant Mendelian randomization studies. To provide more credible evidence on this important and contested public health question, we conducted a Mendelian randomization study to assess the impact of urinary sodium on overall stroke and ischemic stroke, atrial fibrillation, and heart failure, and cardiovascular risk factors using relevant genome wide association studies (GWAS) [13]. We also assessed the impact on established cardiovascular risk factors (e.g. glycemic traits and lipids) as they may provide mechanistic insights.

2. Material and methods

This is a two-sample Mendelian randomization study using publicly available GWAS, which has three main assumptions. First, the genetic instruments should be strongly and independently associated with sodium, here taken as a genome wide significant association (p-value$<5 \times 10^{-8}$) with $r^2 < 0.001$. Second, the association of the genetic instruments with the outcomes should not be confounded. Third, the relation of genetic instruments with the outcome should only be via sodium, i.e., the exclusion restriction assumption [13,16]. We considered whether this assumption might be violated by horizontal pleiotropy where the instruments affect the outcomes directly or via other well-established causes of CVD that affect urinary sodium [17], or by selection bias from surviving of the exposure, particularly when competing risk of the outcome exists [18].

2.1. Ethics approval

The MR study only uses published or publicly-available data. No original data were collected for the MR study. Ethical approval for each of the studies included in the investigation can be found in the original publications (including informed consent from each participant).

2.2. Genetic predictors of urinary sodium

Genetic predictors of urinary sodium were identified based on a published GWAS of the UK Biobank, which consisted of approximately 500,000 participants in the Great Britain [14,19]. The median urinary sodium was 68.4 mmol/L. The GWAS was restricted to participants of European descent, as determined by self-report and principal components analysis clustering, giving up to 446,237 participants. Urinary sodium from spot urine samples was measured by the ion selective electrode method (potentiometric method) using Beckman Coulter AU5400, UK Ltd. Imputation was based on the Haplotype Reference Consortium panel. Genetic associations with log transformed urinary sodium were obtained using a linear mixed model which controlled for population stratification and relatedness amongst participants, and was adjusted for age and sex [14]. The GWAS identified 51 lead single nucleotide polymorphisms (SNPs) associated with urinary sodium. The genes were related to a variety of functions such as congenital anomalies of kidney and urinary tract, weight loss, thermoregulation, learning, behaviour and cognition.

2.3. Genetic associations with outcomes

Complete summary GWAS statistics for the outcomes stroke and its subtypes (MEGASTROKE consortium) [20], atrial fibrillation [21], heart failure (HERMES consortium) [22], LDL, HDL cholesterol and triglycerides (GLGC) [23], type 2 diabetes (DIAGRAM) [24], fasting glucose, insulin, and glycated hemoglobin (MAGIC) [25,26], were obtained from publicly available online GWAS summary data repositories, with majority of them retrieved via MR Base [27]. Table 1 summarizes the numbers (including the cases and controls where relevant) included in these GWAS, population (including ethnicity) and the sample size in the GWAS. Additional details, including quality control, imputation methods, and any covariates adjusted for in each GWAS and how the outcomes were defined are provided in Supplemental Table 1.

2.4. Exploring possible sources of horizontal pleiotropy

We also assessed the association of the genetic instruments with possible sources of horizontal pleiotropy which can lead to violation of InSIDE assumption, i.e., specifically here factors predicted by the instruments that cause both urinary sodium and CVD.

Table 1
Summary of the genome wide association studies included in this Mendelian randomization study.

Exposure	Data source (PMID)	Sample size (% cases)	% European	% Overlap with UK Biobank
Urinary sodium (log transformed)	UK Biobank (31409800)	446,237	100	100
Outcomes	**Data source (PMID)**	**Sample size (% cases)**	**% European**	
Stroke	MEGASTROKE (29531354)	446,696 (9%)	100	0
Ischemic stroke	MEGASTROKE (29531354)	440,328 (8%)	100	0
Cardioembolic stroke	MEGASTROKE (29531354)	211,763 (3%)	100	0
Small vessel stroke	MEGASTROKE (29531354)	198,048 (3%)	100	0
Large artery stroke	MEGASTROKE (29531354)	150,765 (3%)	100	0
Atrial fibrillation	PMID: 30061737	1,030,836 (6%)	100	38
Heart failure	HERMES (31919418)	977,320 (5%)	100	40
LDL cholesterol (SD)	GLGC (24097068)	173,082	100	0
HDL cholesterol (SD)	GLGC (24097068)	187,167	100	0
Triglycerides (SD)	GLGC (24097068)	177,861	100	0
Glucose (mmol/L)	MAGIC (22581228)	58,074	100	0
HbA1c (%)	MAGIC (28898252)	123,665	100	0
Insulin (log)	MAGIC (22581228)	51,750	100	0
Type 2 diabetes	DIAGRAM (28566273)	159,208 (17%)	100	0
Confounders	**Data source (PMID)**	**Sample size**	**% European**	
Body mass index (SD)	GIANT (25673413)	339,224	95	0
Years education attained (SD)	SSGAC (27225129)	328,917	100	34
Alcohol (SD of Log transformed drinks per week)	GSCAN (30642351)	941,280	100	33
Smoking Heaviness (SD of cigarettes per day)	GSCAN (30642351)	337,334	100	36
Estimated glomerular filtration rate (ml/min per 1.73m^2)	COGENT-Kidney Consortium (30604766)	81,829	29	0

These included education (SSGAC) [28], body mass index (BMI) (GIANT) [29], smoking and alcohol use (GSCAN) [30]. We also included renal function (COGENT-Kidney consortium) given kidney function relates strongly with urinary sodium and may cause CVD [31]. Details of these GWAS can also be found in Table 1 and Supplemental Table 1.

2.5. Exposure

The exposure was log transformed urinary sodium.

2.6. Outcomes

The primary outcomes were all stroke and ischemic stroke, atrial fibrillation, and heart failure. The secondary outcomes were ischemic stroke subtypes, LDL and HDL cholesterol (SD), triglycerides (SD), fasting glucose (mmol/L), glycated hemoglobin (%), insulin (log) and type 2 diabetes.

2.7. Allele harmonization

We combined the genetic information from the urinary sodium GWAS and the cardiovascular outcomes GWAS by aligning the effect alleles and the corresponding estimate direction. We also used effect allele frequency to ensure palindromic instruments are aligned properly. For genetic instruments not available for an outcome, a proxy instrument in high LD with the original instrument ($r^2 > 0.8$) was identified via MR-Base based on 1000 Genomes catalog. No proxy instruments were identified for outcomes not available in MR-Base.

2.8. Statistical analysis

We approximated the r^2 of each instrument and calculated the overall F statistic [32,33]. We calculated the Wald ratio of each instrument (SNP on outcome divided by SNP on exposure), i.e., the causal effect, using inverse variance weighting (IVW) with multiplicative random effects [34]. We also estimated the heterogeneity of Wald ratios using the Cochrane Q test, where a significant test indicates the presence of potentially invalid SNPs. IVW assumes balanced pleiotropy and the instrument strength independent of direct effect (InSIDE) assumption, i.e. the instrument does not act via confounders of exposure on outcome, although it is empirically difficult to test these assumptions. We also conducted complementary sensitivity analyses, which rely on different assumptions to assess the robustness of the findings. Triangulating the evidence by checking for consistency across analyses with different assumptions may improve certainty of the results [35].

2.9. Weighted median

The weighted median gives a valid estimate as long as 50% of the weight is derived from valid instruments [36].

2.10. Contamination mixture method

The contamination mixture method used a likelihood function based on estimates from each instrument [37]. Specifically, it assumes the effect of valid instruments is normally distributed around the causal effect whilst the effect of invalid instruments normally distributed around 0 with a large standard deviation. The causal effect was then computed using a profile likelihood by identifying valid and invalid instruments which maximized the likelihood for a particular causal effect [37]. We used the default setting, i.e. 1.5 times the standard deviation of the ratio estimates, for the standard deviation of the distribution of invalid estimands (psi) although we also assessed the impact of other psi values (70%−130% of the default psi) on the estimates.

2.11. Mendelian randomization pleiotropy residual sum and outlier (MR-PRESSO) test

MR-PRESSO provides another statistical means of detecting biases due to pleiotropy (global test), as well as providing a corrected estimate via outlier removal [38]. In brief, MR-PRESSO corrects for horizontal pleiotropy via outlier removal. MR-PRESSO requires the InSIDE assumption, balanced pleiotropy, and that at least 50% of the SNPs are valid instruments [38]. The MR-PRESSO also provides a distortion test to explore whether the analyses with or without outliers produced similar estimates.

2.12. Multivariable Mendelian randomization

Multivariable Mendelian randomization was used to adjust for violations of the InSIDE assumption by horizontal pleiotropy, i.e., where the genetic instruments for urinary sodium affect potential confounders of urinary sodium on the outcome. Education, BMI, smoking, alcohol and renal function were considered as potential confounders. Whether urinary sodium was a cause of these potential confounders (horizontal pleiotropy) or a consequence of these potential confounders (vertical pleiotropy) was identified using bi-directional Mendelian randomization [17]. Potential confounders were considered to be horizontally pleiotropic if they were the result of the urinary sodium instruments and also caused urinary sodium [17]. In the multivariable Mendelian randomization, we calculated the conditional F statistics to assess weak instrument bias, assuming covariance between genetic instruments on each exposure was 0 [39].

2.13. Power calculations

We calculated power for each analysis using the approximation that the sample size for a Mendelian randomization study is the sample size for exposure on outcome divided by the r^2 for genetic instruments on exposure [40].

All analyses were performed using R Version 3.6.1 (R Development Core Team, Vienna, Austria) using R packages ("TwoSampleMR") [27], ("MendelianRandomization") [41], and ("MRPRESSO") [38].

3. Results

Of the 51 SNPs predicting sodium, 8 correlated SNPs were excluded ($r^2 \geq 0.001$), giving 43 SNPs retained in the analyses (Supplemental Table 2). The F statistic for each SNP was greater than 10, with an overall F statistic of 49. The total r^2 for genetic instruments on sodium was 0.0047, so our study had 80% power with 5% alpha to detect an odds ratio (OR) per standard deviation of log transformed sodium of 1.25 for all stroke and heart failure, 1.30 for ischemic stroke, and 1.20 for atrial fibrillation. We found that the effect alleles for the urinary sodium instruments (aligned to increased urinary sodium) were associated with lower education attainment and alcohol use, but not with body mass index, eGFR or tobacco use (Supplemental Table 3). Bi-directional Mendelian randomization suggested urinary sodium was associated with lower alcohol use and education although there were also evidence on reverse causation, as shown in Supplemental Table 4. As such, alcohol use and education were adjusted for in the multivariable Mendelian randomization.

Figure 1 shows the association of urinary sodium with cardiovascular diseases and type 2 diabetes. Urinary sodium was associated with higher risk of overall stroke, ischemic stroke, heart failure, and type 2 diabetes. The Cochrane Q test suggested heterogeneity for heart failure, atrial fibrillation and type 2 diabetes. The weighted median and contamination mixture method gave similar conclusions. Changing the psi values did not alter the conclusion. MR-PRESSO suggested potential horizontal pleiotropy for heart failure, atrial fibrillation and type 2 diabetes based on the global test although exclusion of outliers did not change the conclusion (Supplemental Table 5).

Figure 2 shows the association of urinary sodium with cardiovascular risk factors where there is no clear pattern of association, apart from a positive association with triglycerides. Heterogeneity was high for most outcomes. The weighted median and contamination mixture method also gave similar conclusion, and changing the psi values did not alter the conclusion. MR-PRESSO suggested the presence of such pleiotropy for all outcomes except HbA1c. However, exclusion of outliers did not change the conclusion, except for triglycerides where the estimate was attenuated according to the distortion test (p value $< 10^{-4}$) (Supplemental Table 5).

Results from multivariable Mendelian randomization, adjusted for education and alcohol use, were directionally similar (Supplemental Figs. 1 and 2). The conditional F statistics were >10, suggesting weak instrument bias was unlikely.

4. Discussion

This is one of the first and largest Mendelian randomization studies to examine the impact of urinary sodium on stroke, ischemic stroke, atrial fibrillation, and heart failure, and cardiovascular risk factors. Our study provides potentially more credible evidence that an increase in urinary sodium likely increases the risk of overall, ischemic stroke and heart failure. Given the consistent findings of our study and previous studies [5], our study further supports that a reduction in sodium intake would reduce the burden of cardiovascular disease, and may help clarify the controversies surrounding sodium and cardiovascular health [8]. Our study also suggested a possible detrimental impact of urinary sodium on type 2 diabetes consistent with smaller, observational studies [42].

Our study suggests sodium can increase the risk of overall stroke and ischemic stroke. Potential mechanisms may include the increase in arterial stiffness, vascular endothelial dysfunction and blood pressure [4,14], which all increase ischemic stroke risk [43,44]. The estimates for atrial fibrillation were also suggestive of a detrimental impact of sodium albeit non-significant. Whilst this may indicate the effect of sodium on atrial fibrillation may not be causal, we could not rule out the possibility of selection bias. Specifically, atrial fibrillation occurs at older ages than stroke and CAD, as reflected in the diagnosis age in the relevant GWAS (e.g. mean age of 74 years). Hence, an association of sodium with atrial fibrillation may be biased towards the null by competing risk of death from these diseases [18]. Our study did show a positive link of urinary sodium with heart failure. The underlying pathways may include an increase in left ventricular mass following increased sodium intake [45]. Our study suggested urinary sodium positively related to diabetes risk (Fig. 1), which is consistent with some previous studies [42,46]. Whilst it is unclear why sodium, a non-caloric component in a diet, may impact risk of type 2 diabetes, animal studies suggested that a high salt intake diet may induce endogenous fructose production by activating the aldose reductase pathway in the liver, which in turn may increase the risk of obesity and subsequent type 2 diabetes risk [46]. Nevertheless, it is noted that these associations were not consistent with the findings from the glycemic traits (Fig. 2). The underlying reasons are unclear but it is possible that the exclusion of diabetes cases in the GWAS of glycemic traits may explain these discrepancies.

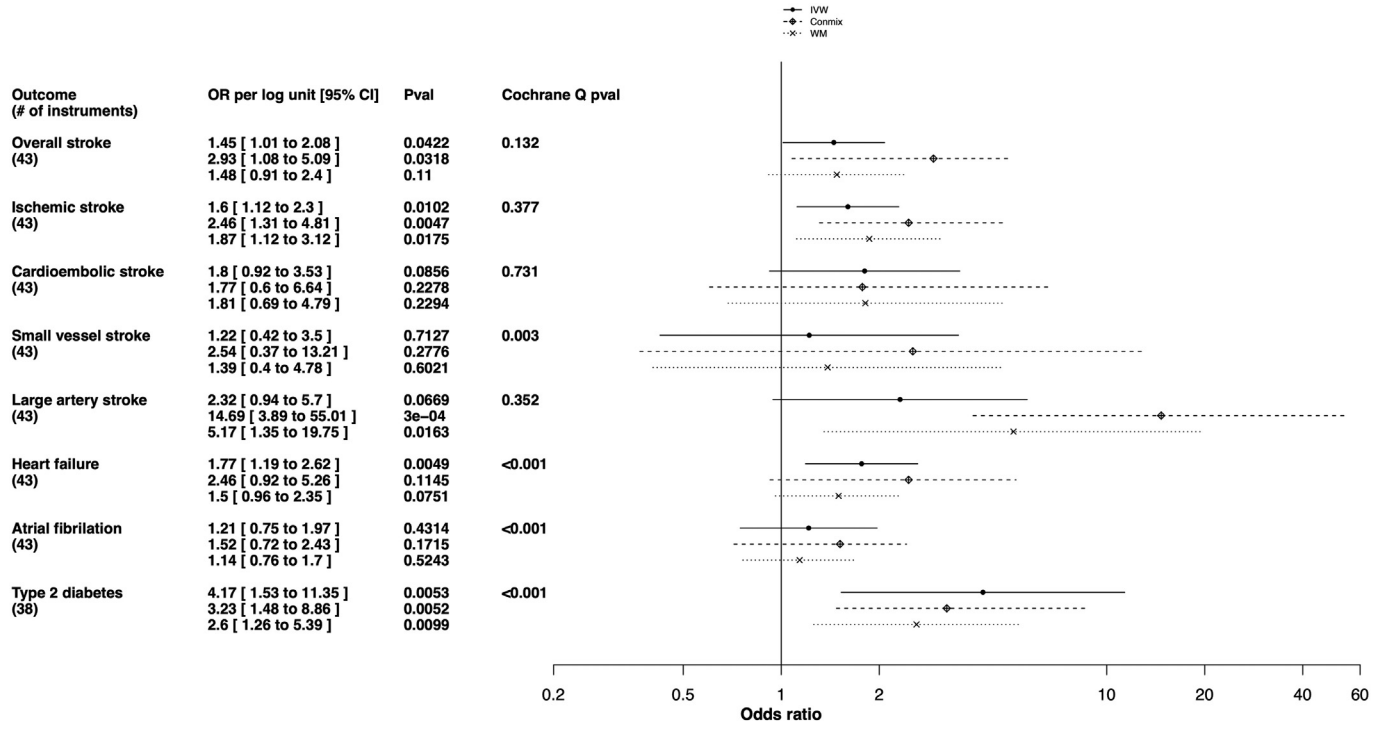

Fig. 1. Impact of urinary sodium on cardiovascular disease and diabetes using Mendelian randomization.

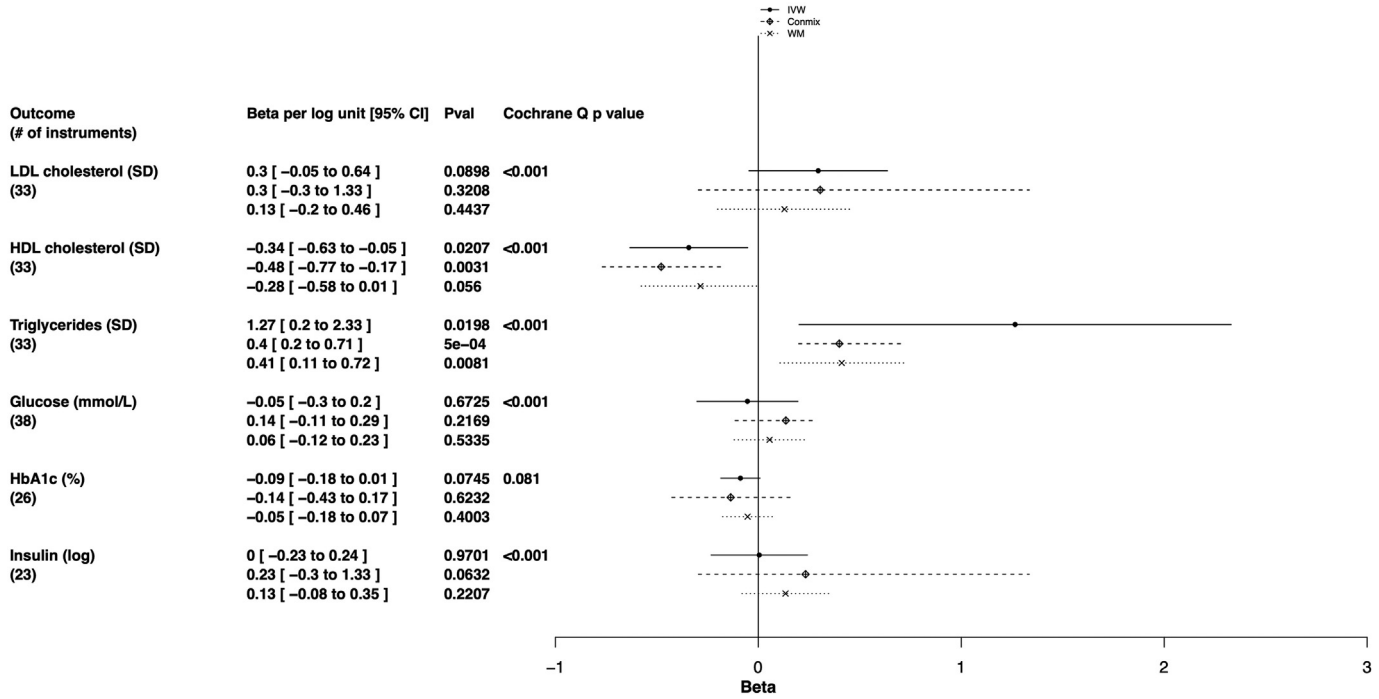

Fig. 2. Impact of urinary sodium on cardiovascular risk factors using Mendelian randomization.

We have not explored the impact of other urinary markers, such as potassium, on cardiovascular risk, as the primary aim of our study was to evaluate the impact of salt intake, using urinary sodium as a proxy. Exploratory analyses suggested urinary potassium, using 13 genetic instruments reported from the potassium GWAS (Supplemental Table 6) [14], was not related to any of the outcomes (Supplemental Figs. 3 and 4) but with very wide confidence intervals, whilst a previous Mendelian randomization study suggested an inverse association with systolic blood pressure [14]. Whether replacing sodium based salt with potassium based salt would be meaningful can be further explored in randomized controlled trials.

The strengths of this study included the use of Mendelian randomization which is more resistant to confounding and the use of large GWAS to reduce the likelihood of false negatives. However, there are some limitations. First, Mendelian randomization relies on three main assumptions, which are difficult to verify empirically. However, we used strong independent genetic instruments for urinary sodium identified from the largest GWAS [14]. Use of genetic instruments likely excludes confounding of instrument on outcome. We used the weighted median, contamination mixture method, and MR-PRESSO, which do not rely on the same set of assumptions about pleiotropy as sensitivity analyses, as well as multivariable Mendelian randomization to rule out the possibility of horizontal pleiotropy via education and alcohol use. In the context of 2 sample MR, we also assumed no overlap between the urinary sodium GWAS and the outcome GWAS, where weak instrument bias would bias our estimates towards null. Whilst this is true for majority of the outcomes (Table 1), there were participant overlaps between the urinary sodium GWAS (UK Biobank) and the atrial fibrillation GWAS (38%) and heart failure GWAS (40%). As such, any weak instrument bias would have biased the estimates towards the confounded exposure-outcome associations. Nevertheless, the F statistics suggested weak instrument bias may not be present. However, these findings could be verified in future studies without the inclusion of UK Biobank participants to assess the impact of participants overlap amongst the exposure and outcome studies. Second, we were not able to assess non-linearity in the relation of log urinary sodium with the outcomes. Future studies using large biobanks may help ascertain the possibility of such relations. Third, we were unable to assess the impact of sodium on hemorrhagic stroke given sufficiently large GWAS summary statistics are not available. The use of smaller GWAS (3226 cases and 3742 non cases) from ISGC gave a very imprecise estimate [47]. This question is particularly important in Asian populations given the higher incidence rate than in Western populations [48], and could be explored in large Asian biobanks. Fourth, spot urine samples may not reflect long-term sodium intake or excretion and hence is subject to misclassification. However, a previous Mendelian randomization study showed the expected relation of urinary sodium with blood pressure, body mass index, and coronary artery disease [14]. Nevertheless, future studies using estimated 24 h urine excretion may help verify our findings. Fifth, we used instruments strongly associated with urinary sodium as a proxy for actual salt intake. We are assuming that higher urinary sodium in the general population is the result of higher salt intake rather than of more efficient salt excretion. This assumption was supported by the correlation of the urinary sodium instruments with salt intake in the UK Biobank, as well as the expected positive association with coronary artery disease and blood pressure [14]. This may also explain why our study may appear contradictory to previous pharmacological studies on sodium/glucose transporter 2 (SGLT2) inhibitors which are potentially cardioprotective despite increased urinary sodium excretion [49]. Specifically, higher urinary sodium in the general population is likely a reflection of higher salt intake in our study whilst higher urinary sodium in the SGLT2 inhibitor studies in patients may reflect the effect of the medication, and hence have generated these differences. Although urinary sodium may also relate to renal function, the urinary sodium SNPs did not strongly predict renal function (Supplemental Table 3). Sixth, we did not use Bonferroni correction since we considered these were outcomes with similar aetiology. However, with correction, the

associations were no longer statistically significant, which could be driven by the generally low statistical power in Mendelian randomization studies. Seventh, we were unable to explore non-linearity of sodium in CVD risk, as suggested previously [50,51], which could be explored in future studies. Finally, our findings may be open to selection bias due to the underlying GWAS inevitably missing people who have already died of the exposure or of competing risk of the outcome. However, any resulting would be towards the null, making our estimates conservative.

5. Conclusion

This Mendelian randomization study suggests urinary sodium likely increases the risk of stroke, heart failure, and type 2 diabetes. Our study further supports public health policies to minimize population sodium intake so as to reduce the disease burden due to cardiovascular diseases.

Sources of funding

This study was partly funded by the Health and Medical Research Fund, Food and Health Bureau, HKSAR Government, Hong Kong, China (CFS-HKU1). The funder had no role in the design, analyses, interpretation of results or writing of the paper.

Author contributions

SLAY and CMS designed the study, wrote the analysis plan and interpreted the results. SLAY undertook analyses with feedback from CMS. SLAY wrote the first draft of the manuscript with critical feedback and revisions from CMS. All authors gave final approval of the version to be published. SLAY had primary responsibility for final content.

Conflict of interest

None.

Acknowledgement

We thank the investigators of the atrial fibrillation GWAS for providing the summary statistics, which have been downloaded from http://csg.sph.umich.edu/willer/public/afib2018/. We thank MEGASTROKE for providing the summary statistics for stroke, which have been downloaded from http://www.megastroke.org/index.html. The MEGASTROKE project received funding from sources specified at http://www.megastroke.org/acknowledgments.html. The list of authors in the MEGASTROKE Consortium can be found in the Supplemental materials. We thank HERMES Consortium for providing the summary statistics for heart failure, which have been downloaded from http://www.broadcvdi.org/informational/data. We thank GLGC for providing summary statistics for lipids, which have been downloaded from http://csg.sph.umich.edu/willer/public/lipids2013/. We thank MAGIC for providing summary statistics for glycemic traits, which have been downloaded from https://www.magicinvestigators.org. We thank DIAGRAM for providing the summary statistics for type 2 diabetes, which have been downloaded from https://www.diagram-consortium.org. We thank GIANT consortium for providing the summary statistics for body mass index, which have been downloaded from https://portals.broadinstitute.org/collaboration/giant/index.php/GIANT_consortium_data_files. We thank SSGAC for providing the summary statistics for years of education attainment, which have been downloaded from https://www.thessgac.org/data. We thank GSCAN for providing the summary statistics for alcohol and cigarette use phenotypes, which have been downloaded from https://conservancy.umn.edu/handle/11299/201564. We thank the investigator of the COGENT-Kidney consortium for providing the summary statistics for eGFR, which have been downloaded from https://www.liverpool.ac.uk/translational-medicine/research/statistical-genetics/data-resources/. We thank the investigator of the urinary sodium GWAS for providing the summary statistics for urinary sodium, which have been downloaded from the GWAS catalog (ebi.ac.uk/gwas/publications/31409800#study_panel). We thank ISGC investigators for providing summary statistics for intracerebral hemorrhage stroke through the ISGC Cerebrovascular Disease Knowledge Portal. We thank Mr KL Wong for technical support.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.clnu.2020.09.018.

References

[1] GBD 2017 Diet Collaborators. Health effects of dietary risks in 195 countries, 1990–2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet 2019;393:1958–72.
[2] Stamler J, Rose G, Stamler R, Elliott P, Dyer A, Marmot M. INTERSALT study findings. Public health and medical care implications. Hypertension 1989;14:570–7.
[3] He FJ, MacGregor GA. Role of salt intake in prevention of cardiovascular disease: controversies and challenges. Nat Rev Cardiol 2018;15:371–7.
[4] Appel LJ, Frohlich ED, Hall JE, Pearson TA, Sacco RL, Seals DR, et al. The importance of population-wide sodium reduction as a means to prevent cardiovascular disease and stroke: a call to action from the American Heart Association. Circulation 2011;123:1138–43.
[5] Cook NR, Cutler JA, Obarzanek E, Buring JE, Rexrode KM, Kumanyika SK, et al. Long term effects of dietary sodium reduction on cardiovascular disease outcomes: observational follow-up of the trials of hypertension prevention (TOHP). BMJ 2007;334:885–8.
[6] Paakko TJW, Perkiomaki JS, Silaste ML, Bloigu R, Huikuri HV, Antero Kesaniemi Y, et al. Dietary sodium intake is associated with long-term risk of new-onset atrial fibrillation. Ann Med 2018;50:694–703.
[7] Mills KT, Chen J, Yang W, Appel LJ, Kusek JW, Alper A, et al. Sodium excretion and the risk of cardiovascular disease in patients with chronic kidney disease. JAMA 2016;315:2200–10.
[8] Trinquart L, Johns DM, Galea S. Why do we think we know what we know? A metaknowledge analysis of the salt controversy. Int J Epidemiol 2016;45:251–60.
[9] National Academies of Sciences Engineering and Medicine (U.S.), Committee to Review the Dietary Reference Intakes for Sodium and Potassium, Stallings VA, Harrison M, Oria M, National Academies of Sciences Engineering and Medicine (U.S.). Food and Nutrition Board, National Academies Press (U.S.). Dietary reference intakes for sodium and potassium. Washington, D.C.: National Academies Press; 2019.
[10] Sacks FM, Svetkey LP, Vollmer WM, Appel LJ, Bray GA, Harsha D, et al. Effects on blood pressure of reduced dietary sodium and the Dietary Approaches to Stop Hypertension (DASH) diet. DASH-Sodium Collaborative Research Group. N Engl J Med 2001;344:3–10.
[11] Harsha DW, Sacks FM, Obarzanek E, Svetkey LP, Lin PH, Bray GA, et al. Effect of dietary sodium intake on blood lipids: results from the DASH-sodium trial. Hypertension 2004;43:393–8.
[12] Chang HY, Hu YW, Yue CS, Wen YW, Yeh WT, Hsu LS, et al. Effect of potassium-enriched salt on cardiovascular mortality and medical expenses of elderly men. Am J Clin Nutr 2006;83:1289–96.
[13] Lawlor DA. Commentary: Two-sample Mendelian randomization: opportunities and challenges. Int J Epidemiol 2016;45:908–15.
[14] Pazoki R, Evangelou E, Mosen-Ansorena D, Pinto RC, Karaman I, Blakeley P, et al. GWAS for urinary sodium and potassium excretion highlights pathways shared with cardiovascular traits. Nat Commun 2019;10:3653.
[15] Zanetti D, Bergman H, Burgess S, Assimes TL, Bhalla V, Ingelsson E. Urinary albumin, sodium, and potassium and cardiovascular outcomes in the UK biobank: observational and Mendelian randomization analyses. Hypertension 2020;75:714–22.
[16] Davies NM, Holmes MV, Davey Smith G. Reading Mendelian randomisation studies: a guide, glossary, and checklist for clinicians. BMJ 2018;362:k601.
[17] Yang Q, Sanderson E, Tilling K, Borges MC, Lawlor DA. Exploring and mitigating potential bias when genetic instrumental variables are associated with multiple non-exposure traits in Mendelian randomization. medRxiv 2019:19009605.

[18] Schooling C, Lopez P, Yang Z, Au Yeung S, Huang J. Bias from competing risk before recruitment in Mendelian Randomization studies of conditions with shared etiology. bioRxiv 2020. https://www.biorxiv.org/content/10.1101/716621v4.
[19] Sudlow C, Gallacher J, Allen N, Beral V, Burton P, Danesh J, et al. UK biobank: an open access resource for identifying the causes of a wide range of complex diseases of middle and old age. PLoS Med 2015;12:e1001779.
[20] Malik R, Chauhan G, Traylor M, Sargurupremraj M, Okada Y, Mishra A, et al. Multiancestry genome-wide association study of 520,000 subjects identifies 32 loci associated with stroke and stroke subtypes. Nat Genet 2018;50:524–37.
[21] Nielsen JB, Thorolfsdottir RB, Fritsche LG, Zhou W, Skov MW, Graham SE, et al. Biobank-driven genomic discovery yields new insight into atrial fibrillation biology. Nat Genet 2018;50:1234–9.
[22] Shah S, Henry A, Roselli C, Lin H, Sveinbjornsson G, Fatemifar G, et al. Genome-wide association and Mendelian randomisation analysis provide insights into the pathogenesis of heart failure. Nat Commun 2020;11:163.
[23] Global Lipids Genetics Consortium, Willer CJ, Schmidt EM, Sengupta S, Peloso GM, Gustafsson S, et al. Discovery and refinement of loci associated with lipid levels. Nat Genet 2013;45:1274–83.
[24] Scott RA, Scott LJ, Magi R, Marullo L, Gaulton KJ, Kaakinen M, et al. An expanded genome-wide association study of type 2 diabetes in Europeans. Diabetes 2017.
[25] Manning AK, Hivert MF, Scott RA, Grimsby JL, Bouatia-Naji N, Chen H, et al. A genome-wide approach accounting for body mass index identifies genetic variants influencing fasting glycemic traits and insulin resistance. Nat Genet 2012;44:659–69.
[26] Wheeler E, Leong A, Liu CT, Hivert MF, Strawbridge RJ, Podmore C, et al. Impact of common genetic determinants of Hemoglobin A1c on type 2 diabetes risk and diagnosis in ancestrally diverse populations: a transethnic genome-wide meta-analysis. PLoS Med 2017;14:e1002383.
[27] Hemani G, Zheng J, Elsworth B, Wade KH, Haberland V, Baird D, et al. The MR-base platform supports systematic causal inference across the human phenome. Elife 2018;7.
[28] Okbay A, Beauchamp JP, Fontana MA, Lee JJ, Pers TH, Rietveld CA, et al. Genome-wide association study identifies 74 loci associated with educational attainment. Nature 2016;533:539–42.
[29] Locke AE, Kahali B, Berndt SI, Justice AE, Pers TH, Day FR, et al. Genetic studies of body mass index yield new insights for obesity biology. Nature 2015;518:197–206.
[30] Liu M, Jiang Y, Wedow R, Li Y, Brazel DM, Chen F, et al. Association studies of up to 1.2 million individuals yield new insights into the genetic etiology of tobacco and alcohol use. Nat Genet 2019;51:237–44.
[31] Morris AP, Le TH, Wu H, Akbarov A, van der Most PJ, Hemani G, et al. Trans-ethnic kidney function association study reveals putative causal genes and effects on kidney-specific disease aetiologies. Nat Commun 2019;10:29.
[32] Burgess S, Thompson SG. Avoiding bias from weak instruments in Mendelian randomization studies. Int J Epidemiol 2011;40:755–64.
[33] Pierce BL, Ahsan H, Vanderweele TJ. Power and instrument strength requirements for Mendelian randomization studies using multiple genetic variants. Int J Epidemiol 2011;40:740–52.
[34] Bowden J, Del Greco MF, Minelli C, Davey Smith G, Sheehan N, Thompson J. A framework for the investigation of pleiotropy in two-sample summary data Mendelian randomization. Stat Med 2017;36:1783–802.
[35] Au Yeung SL, Borges MC, Lawlor DA. Association of genetic instrumental variables for lung function on coronary artery disease risk: a 2-sample Mendelian randomization study. Circ Genom Precis Med 2018;11:e001952.
[36] Bowden J, Davey Smith G, Haycock PC, Burgess S. Consistent estimation in Mendelian randomization with some invalid instruments using a weighted median estimator. Genet Epidemiol 2016;40:304–14.
[37] Burgess S, Foley CN, Allara E, Staley JR, Howson JMM. A robust and efficient method for Mendelian randomization with hundreds of genetic variants. Nat Commun 2020;11:376.
[38] Verbanck M, Chen CY, Neale B, Do R. Detection of widespread horizontal pleiotropy in causal relationships inferred from Mendelian randomization between complex traits and diseases. Nat Genet 2018;50:693–8.
[39] Sanderson E, Davey Smith G, Windmeijer F, Bowden J. An examination of multivariable Mendelian randomization in the single-sample and two-sample summary data settings. Int J Epidemiol 2019;48:713–27.
[40] Freeman G, Cowling BJ, Schooling CM. Power and sample size calculations for Mendelian randomization studies using one genetic instrument. Int J Epidemiol 2013;42:1157–63.
[41] Yavorska OO, Burgess S. Mendelian randomization: an R package for performing Mendelian randomization analyses using summarized data. Int J Epidemiol 2017;46:1734–9.
[42] Radzeviciene L, Ostrauskas R. Adding salt to meals as a risk factor of type 2 diabetes mellitus: a case-control study. Nutrients 2017;9.
[43] Balkaya M, Prinz V, Custodis F, Gertz K, Kronenberg G, Kroeber J, et al. Stress worsens endothelial function and ischemic stroke via glucocorticoids. Stroke 2011;42:3258–64.
[44] Tuttolomondo A, Di Sciacca R, Di Raimondo D, Serio A, D'Aguanno G, Pinto A, et al. Arterial stiffness indexes in acute ischemic stroke: relationship with stroke subtype. Atherosclerosis 2010;211:187–94.
[45] Ferrara LA, de Simone G, Pasanisi F, Mancini M, Mancini M. Left ventricular mass reduction during salt depletion in arterial hypertension. Hypertension 1984;6:755–9.
[46] Lanaspa MA, Kuwabara M, Andres-Hernando A, Li N, Cicerchi C, Jensen T, et al. High salt intake causes leptin resistance and obesity in mice by stimulating endogenous fructose production and metabolism. Proc Natl Acad Sci U S A 2018;115:3138–43.
[47] Woo D, Falcone GJ, Devan WJ, Brown WM, Biffi A, Howard TD, et al. Meta-analysis of genome-wide association studies identifies 1q22 as a susceptibility locus for intracerebral hemorrhage. Am J Hum Genet 2014;94:511–21.
[48] Tsai CF, Thomas B, Sudlow CL. Epidemiology of stroke and its subtypes in Chinese vs white populations: a systematic review. Neurology 2013;81:264–72.
[49] Cianciolo G, De Pascalis A, Capelli I, Gasperoni L, Di Lullo L, Bellasi A, et al. Mineral and electrolyte disorders with SGLT2i therapy. JBMR Plus 2019;3:e10242.
[50] O'Donnell M, Mente A, Rangarajan S, McQueen MJ, O'Leary N, Yin L, et al. Joint association of urinary sodium and potassium excretion with cardiovascular events and mortality: prospective cohort study. BMJ 2019;364:l772.
[51] Mente A, O'Donnell M, Rangarajan S, McQueen M, Dagenais G, Wielgosz A, et al. Urinary sodium excretion, blood pressure, cardiovascular disease, and mortality: a community-level prospective epidemiological cohort study. Lancet 2018;392:496–506.