**Author’s response to reviews**

**Title:** Estimating mean population salt intake in Fiji and Samoa using spot urine samples

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**REVIEWER 1**

**COMMENT:** The results are promising and the information could be interesting for the readers. Just two minor comments:

- The BMI is quiet high for a non-selected population in that area. Do the authors have any information about the energy intake, related to the salt intake, obviously?

**RESPONSE:** Thank you for your comment. We agree that the measured BMI from the sample in Fiji and Samoa is high. This is in line with previous reports that show that the Pacific region has some of the highest rates of overweight and obesity globally [reference: World Bank. Non-Communicable Disease (NCD) Roadmap Report. Washington, DC: World Bank Group; 2014]. There was no available data on energy intake from the GACD Pacific Salt Project (the source of
the data for the secondary analyses), hence, we were unable to measure the relationship of energy and salt intake.

CHANGES MADE TO THE MANUSCRIPT:

On Discussion, Lines 302-303, we added the phrase: “The mean BMI in Fiji and Samoa is much higher (29 and 33kg/m2, respectively), which is in line with previous reports that show that the Pacific region has some of the highest rates of overweight and obesity globally.”

COMMENT: - sodium excretion is definitely influenced by the use of diuretics. The authors should provide additional information in this field.

RESPONSE: Thank you for your comment. We agree that sodium excretion is influenced by the use of diuretics. Sodium excretion is likely to be higher in the few hours after a diuretic is taken, which may lead to overestimation of salt intake. In the GACD Pacific Salt Project, however, only a small number of participants in both countries reported use of medications to lower blood pressure. In addition, we also think that this is less relevant to our secondary analyses, given that the main objective was to evaluate the agreement between spot urine and 24-hour urine samples in estimating population-level salt intake (where the spot urine sample was collected as part of the 24-hour urine collection), rather than providing nationally representative salt intake estimates.

REVIEWER 2

COMMENT: This study compares 24 hour salt level using spot urine and 24-hour urine collection in Fiji and Samoa. The strength of this study is the large sample size, testing five equations to convert spot urine to 24-hour urine sodium estimation. As expected, there was a wide variation according to the methods used for conversion. All pairwise comparisons of salt intake estimates based on spot urine equations against 24-hour urine values were significantly different which is creating even more uncertainty for using spot urine method. The authors suggested to have additional analyses but did not explain what that would be. Do the authors mean we need more studies like this or more analyses? If more analyses, what could be they?

RESPONSE: Thank you for your comments. We meant that more studies where both 24-hour urine and spot urine samples are collected are needed to further assess the capacity of spot urine samples to estimate mean salt intake in both countries. To the best of our knowledge, this is only the first study to evaluate the use of spot urine samples in Fiji and Samoa, thus, more studies are required before they can be confidently used in these populations.

CHANGES MADE TO THE MANUSCRIPT:
On Abstract, Lines 71-73, we clarified: “These data suggest that additional studies where both 24-hour urine and spot urine samples are collected are needed to further assess whether methods based on spot urine samples can be confidently used to estimate mean population salt intake in Fiji and Samoa.”

On Discussion, Lines 287-290, we rephrased and clarified: “More robust studies where both 24-hour urine and spot urine samples are collected are required to fully understand the capacity of spot urine samples to estimate mean salt intake in Fiji and Samoa, before they can be confidently used in these populations.”

COMMENT: The International Consortium for Quality Research on Dietary Sodium/Salt (TRUE) recommends simultaneous calibration study with complete 24-hour urine samples should ideally be conducted in an adequately large subset to ensure the accuracy of the spot urine estimate. What are the thoughts of authors on this?

RESPONSE: Thank you for your question. We agree that this is important. As above, we believe that more robust studies are needed to fully understand the capacity of spot urine samples to estimate population-level salt intake in different population groups; hence, it is essential to continue to collect complete 24-hour urine samples in an adequately large subset, so we can validate the estimates from spot urine samples.

CHANGES MADE TO THE MANUSCRIPT:

On Discussion, Lines 350-352, we added: “It is therefore important to continue to collect both 24-hour urine and spot urine samples in adequately large samples, so that the salt intake estimates from spot urine samples (using different equations) can be validated.”

COMMENT: Further, the INTERSALT equation gave the best estimation for both Fiji and Samoa. It may be useful for the readers if authors can add why INTERSALT showed the closest estimation compared to other equations. What could be the reasons (body mass index, male/female ratio, age, ethnicity etc.) for such a close estimation?

RESPONSE: It is not possible to ascertain which element of the INTERSALT equation (body mass index, sex, age, etc) is responsible for the close estimation between spot and 24-urine samples. Nevertheless, we added information in the manuscript to provide the readers a brief background about the equations.

CHANGES MADE TO THE MANUSCRIPT:

On Methods, Lines 169-173, we added: “The INTERSALT equation is based on a regression model that includes age, sex, BMI, and spot urine measurements (spot sodium, creatinine, with
or without potassium). The other equations estimate 24-hour sodium excretion by adjusting the ratio of spot urine sodium and creatinine by the predicted 24-hour creatinine excretion.”

On Discussion, Line 279-280, we rephrased: “…despite it being originally developed using data from 29 populations (large sample of more than 5000 participants) in North America and Europe.”

COMMENT: One of the limitations of the study which is missing in the manuscript is that the spot urine was part of 24-hour urine and this could inflate the correlation between the two collections.

RESPONSE: Thank you for your comment. We previously mentioned this under the strengths and limitations section of the Discussion, Lines 370-371.

COMMENT: Regardless of the limitations of the spot urine samples, the authors recommend that spot urine can be used to classify whether population intake at the population level is higher than WHO recommended level or not (>5 g/day). I think this is a very important finding.

RESPONSE: Thank you for your comment. We agree that this is an important finding. This is in line with the systematic review and meta-analysis conducted in 2016 [reference: Huang et al. Mean population salt intake estimated from 24-hour urine samples and spot urine samples: a systematic review and meta-analysis. International Journal of Epidemiology. 2016;45(1):239-50]

COMMENT: The high difference between the amount of salt consumption in Fiji and Samoa is very interesting and that might have an effect on spot urine estimation.

RESPONSE: Thank you for highlighting this. We agree that this is an important point, and have included this in the Discussion, Lines 280-287.

COMMENT: The authors mentioned that they applied the level creatinine excretion for the first time as exclusion criteria in the analyses. What is the rationale of selecting these cut-off points? Are you assuming that the concentration of creatinine throughout the day would be constant?

RESPONSE: Thank you for your question. To clarify, we applied exclusion criteria for both 24-hour urine and spot urine samples. For 24-hour urine samples, we adapted the creatinine excretion (mmol/day) cut-offs used in previous studies to assess the completeness of 24-hour urine collection. This was reported in the Methods, Lines 151-153.
For spot urine samples, we used reference range values for creatinine concentration (mmol/L) to exclude implausible values. To our knowledge, we are the first to apply an exclusion criteria to exclude spot urine samples. The criteria was based on a review of different cut-off points employed by various laboratories. We understand that creatinine excretion is highly variable and could be affected by several factors, which is why we used the widest range available.

CHANGES MADE TO THE MANUSCRIPT:

On Methods, Lines 156-157, we added: “These criteria were based on a review of cut-offs employed by various laboratories. Since creatinine excretion is highly variable and can be affected by numerous factors including age, sex, body mass, ethnicity, exercise, and recent diet, among others, the widest range was used in this study.”

MINOR COMMENTS

COMMENT: Line 97- For these reasons, its application is often limited in population surveys. I do not get this. The purpose of spot urine is also to estimated salt intake at the population level, is not it?

RESPONSE: Thank you for your question. We meant that 24-hour urine collection is often limited in population surveys, since the method is expensive, and can be burdensome to participants due to the complex nature of collection.

CHANGES MADE TO THE MANUSCRIPT:

On Background, Lines 95-98, we rephrased: “However, the use of 24-hour urine collection is limited in some population surveys, since the method is expensive, and can be burdensome to participants due to the complex nature of collection. This often results in poor participation rates and incomplete urine samples.”

COMMENT: There is a growing interest in the use of spot urine samples as an alternative to the gold standard 24-hour urine collection in measuring population-level salt intake. My impression is that over the last few years, there is a decreasing interest in the use of spot urine samples as an alternative to 24-hour urine collection. What made the authors think that there is an increasing interest to use spot urine? Definitely, there is an increasing interest to find a more convenient and easy way to measuring sodium intake than 24-hour urine collection.

RESPONSE: We agree that there is a growing interest in finding less costly and burdensome alternatives to 24-hour urine collection, and we cited spot urine samples as an example in Line 100. In the past years, we found a number of studies (in countries including Viet Nam, India, Australia, New Zealand, US, Vanuatu, among others) assessing the capacity of the different spot
equations to measure population-level salt intake. In addition, some countries have incorporated salt intake measurement through spot urine collection in their STEPS survey.

CHANGES MADE TO THE MANUSCRIPT:

On Abstract, Lines 54-55, we rephrased: “There is an increasing interest in finding less costly and burdensome alternatives to measuring population-level salt intake than 24-hour urine collection, such as spot urine samples.”

COMMENT: Line 126 …..evaluate the change in salt intake after 18-20 months of salt reduction interventions. Does this mean this is the salt estimation in the area where intervention was implemented?

RESPONSE: Thank you for your question. The GACD Pacific Salt Project in Fiji and Samoa was implemented at a national level, and the samples at baseline and follow-up (for each country) were selected through nationally representative cross-sectional surveys. For these secondary analyses, we combined the baseline and follow-up data points for each country. This was possible since the data were collected from different individuals at each time point and are therefore independent. We reported this in the Methods, Lines 193-195.

COMMENT: Line 131: What about menstruating women?

RESPONSE: Thank you for highlighting this. Yes, we also excluded menstruating women.

CHANGES MADE TO THE MANUSCRIPT:

On Methods, Line 133, we added: “Participants who were pregnant, lactating, or menstruating at the time of the survey were excluded.”

COMMENT: Line 152 : total creatinine excretion was <4mmol/day or >25mmol/day for women, and <6mmol/day or >30mmol/day for men. Something is wrong with the unit.

RESPONSE: Thank you for your comment. We’ve checked this, and believe that this is the correct unit. We’ve calculated creatinine excretion (mmol/day) by multiplying the creatinine concentration (mmol/L) from the 24-hour urine samples by the 24-hour urine volume (L/day).

COMMENT: Line 196: starting with number. I think the journal does no support sentence starting with the number.
RESPONSE: Thank you for highlighting this. We’ve changed the figure (number) to words.

CHANGES MADE TO THE MANUSCRIPT:

On Results, Line 203, we changed: “Six hundred sixty nine and 998 individuals in Fiji and Samoa…”

COMMENT: 319 This may be related to the smaller sample size in Fiji, which led to higher variability in the sample. The sample size should not be the cause of the difference as long as it is adequately powered.

RESPONSE: Thank you for your comment. We agree, and removed this sentence from the manuscript.

COMMENT: 345 This shows that while spot urines may be flawed for individual assessment of intake, this method can still produce reasonable population-level estimates. The results of the study do not strongly support this statement.

RESPONSE: Thank you for this comment. To clarify, this line refers to our assessment of the Bland-Altman plots, which show that apart from the Kawasaki and Mage equations, the mean bias line in the plots from the other equations (especially the INTERSALT with K equation) is reasonably close to zero, which means that these equations produced ‘reasonable’ population-level salt intake estimates (i.e. estimates that are close to the mean 24-hour urine value). However, we agree that we might have over-interpreted this, so we rephrased the sentence, and made minor changes throughout the manuscript to reflect this.

CHANGES MADE TO THE MANUSCRIPT:

On Discussion, Lines 348-353, we rephrased: “This suggests that while methods based on spot urines may be flawed for individual assessment of intake, they might be able to provide estimates of population-level salt intake that are close to the 24-hour urine estimates. It is therefore important to continue to collect both 24-hour urine and spot urine in adequately large samples, so that the salt intake estimates from spot urine samples (using different equations) can be validated.”

In addition, minor changes were made to reflect the above interpretation of our results:

On Abstract, Lines 71-73: “These data suggest that additional studies where both 24-hour urine and spot urine samples are collected are needed to further assess whether methods based on spot urine samples can be confidently used to estimate mean population salt intake in Fiji and Samoa.”
On Discussion, Lines 287-290: “More robust studies where both 24-hour urine and spot urine samples are collected are required to fully understand the capacity of spot urine samples to estimate mean salt intake in Fiji and Samoa, before they can be confidently used in these populations.”

On Conclusion, Lines 378-379: “These data suggest that additional studies are needed before spot urine samples can be confidently used to estimate population-level salt intake in Fiji and Samoa.”

On Conclusion, Lines 382-384: “More studies where both 24-hour urine and spot urine samples are collected are required to further evaluate the capacity of spot urines to estimate 24-hour salt intake in these populations.”

COMMENT: I think the journal follows abbreviated name of journals.
RESPONSE: Thank you for this suggestion. We’ve abbreviated the name of the Journals in the References.

COMMENT: I would keep either Table 2 or Forest Plot. Both are giving the same information.
RESPONSE: Thank you for this suggestion. We’ve kept the figure and removed the Table 2.