Author’s response to reviews

Title: Low-moderate arsenic exposure and respiratory health in American Indian communities in the Strong Heart Study

Authors:
Martha Powers (mpower24@jhu.edu)
Tiffany Sanchez (trs2111@cumc.columbia.edu)
Maria Grau-Perez (maria.grau.perez@gmail.com)
Fawn Yeh (Fawn-Yeh@ouhsc.edu)
Kevin Francesconi (kevin.francesconi@uni-graz.at)
Walter Goessler (walter.goessler@uni-graz.at)
Christine George (cgeorg19@jhu.edu)
Christopher Heaney (cheaney1@jhu.edu)
Lyle Best (lbest@restel.com)
Jason Umans (jason.umans@gmail.com)
Robert Brown (rbrown@jhmi.edu)
Ana Navas-Acien (an2737@cumc.columbia.edu)

Version: 1 Date: 10 Sep 2019

Author’s response to reviews:

ENHE-D-19-00137
Low-moderate arsenic exposure and respiratory health in American Indian communities in the Strong Heart Study

Martha Powers; Tiffany R Sanchez, PhD; Maria Grau-Perez, MS; Fawn Yeh, PhD; Kevin Francesconi, PhD; Walter Goessler, PhD; Christine M. George, PhD; Christopher Heaney, PhD; Lyle G. Best, MD; Jason G. Umans, MD, PhD; Robert H. Brown, MD; Ana Navas-Acien, MD, PhD

Reviewer Comments:
Reviewer #1: The authors present an interesting study of how arsenic exposure is associated with respiratory function among a sample of different American Indian communities. They found that higher concentrations of arsenic were associated with a number of indicators of respiratory impairments, including obstructive/restrictive patterns, as well as decreased function (FEV1 & FVC). This is well-written, and the data support the authors' conclusions. This is an important and well-done study, performed in an underrepresented population, that would be of great interest to the readers of Environmental Health. I would recommend it for publication and only have minor suggested revisions:

Comment: Methods/Results: did the authors explore whether the different American Indian communities had similar exposure-response patterns either through testing for an interaction or stratified analyses? It would be interesting to know if the responses are similar across those different communities which may have differing sources of As exposure, different potential confounding structures, and/or different susceptibilities to As effects on respiratory outcomes.

Response: We examined for potential interaction by study site, but these analyses were ultimately not included in the paper (please see table below). We did not see statistical evidence of interaction when examining airflow obstruction and restrictive pattern by study location. In our study population, arsenic exposure is closely related to site, as arsenic levels in drinking water are higher in Arizona, lower in Oklahoma, and intermediate in North and South Dakota. Despite these differences, the levels still overlap across sites, what allowed us to adjust in regression models. In model 1, adjustment for study site resulted in little change in the measures of association, suggesting that the findings are robust.

Weighted Odds Ratios (95% Confidence Interval) for Airflow Obstruction and Restrictive Pattern, Defined Based on Fixed Ratios, when an Interquartile Range* of Urinary Arsenic Concentration is Compared, by Participant Study Site:

<table>
<thead>
<tr>
<th>Variable</th>
<th>Airflow obstruction</th>
<th></th>
<th>Restrictive pattern</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N</td>
<td>Odds Ratio 95% CI</td>
<td>P for interaction</td>
<td>N</td>
</tr>
<tr>
<td>Study site</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AZ</td>
<td>226</td>
<td>1.11</td>
<td>(0.50, 2.44)</td>
<td>258</td>
</tr>
<tr>
<td>OK</td>
<td>770</td>
<td>1.32</td>
<td>(1.01, 1.72)</td>
<td>779</td>
</tr>
<tr>
<td>SD/ND</td>
<td>829</td>
<td>1.12</td>
<td>(1.01, 1.36)</td>
<td>637</td>
</tr>
</tbody>
</table>

Models adjusted for sex, age, education, smoking status, cigarette pack-years, body mass index, diabetes, estimated glomerular filtration rate, and tuberculosis.
*Interquartile range of the sum inorganic and methylated urinary arsenic concentrations was 5.8 to 16.7 µg/g creatinine.

ORs were stratified by each subgroup of interest, and associated P values for interaction were obtained from models with interaction terms and using Wald tests for multiple coefficients.

Comment: Lines 132: please clarify if the self-reported respiratory diagnoses include prior diagnoses,
current, or both.

Response: We have updated this sentence to now read: “Participants self-reported if a medical person ever told them that they had emphysema, asthma, or chronic bronchitis.”

Comment: Lines 182-184: Rather than just saying "full adjustment", say what as adjusted for here.

Response: We have updated the sentence to include the covariates: “After full adjustment (age, sex, education, site, smoking status, smoking pack-year, eGFR, tuberculosis, and BMI) (Table 3, model 3), the odds ratio [95% CI] comparing the highest to lowest arsenic tertile (≥14.0 vs. ≤7.0 µg/g creatinine) was 1.33 [0.99, 1.77] for obstruction and 1.34 [0.92, 1.96] for restrictive pattern.”

Comment: Lines 184-186: This statement "comparing interquartile range of arsenic" sounds like a factor variable, but I thought it was a continuous variable. If it is continuous, say "for an interquartile range increase in arsenic".

Response: The sentence has been updated to read: “The corresponding OR [95%CI] for an interquartile range (IQR) increase of arsenic was 1.17 [0.99, 1.38] (P for trend 0.07)...”

Comment: Lines 186-187: Figure 1 is mentioned here, but only briefly, and the text description doesn't adequately describe what is being tested or presented. Please describe these results more thoroughly, and/or reference this figure when you discuss the effect modification results (lines 208-215).

Response: We have further described Figure 1 in the text, to now read: “Modelling urinary arsenic using flexible splines showed positive and linear associations with restrictive pattern and airflow obstruction that were suggestive but nonsignificant in the complete sample.

Comment: The use of splines to assess non-linearity feels very descriptive rather than analytic. Can the authors perform a test as to whether the spline models describe the data better than the IQR-linear models, perhaps a likelihood ratio test?

Response: Yes, we utilized splines for data visualization and descriptive purposes. We have conducted a Wald-test for the non-linear components of the spline model. Those p-values are p=0.08 and 0.11, for the dichotomous outcomes Obstruction and Restrictive pattern, and &lt;0.001 and p=0.005 for FEV1, % predicted and FVC, % predicted, respectively, while the p-value for FEV1/FVC the p-value for non-linearity is 0.53. These findings support a possible non-linear association between arsenic and lung-spirometric outcomes, especially for FEV1 and FVC. We have added these p-values from the spline models to the figure footnotes. We have also added a sentence regarding the possible non-linear association to the Results section.

Comment: Lines 283-285: I also wonder if former smokers have some residual lung tissue damage or immune dysfunction, that would make them more susceptible to toxic effects from As or other risk factors for impaired lung function. Is there any literature supporting this hypothesis? a little more
discussion about why non-smokers might be susceptible to the respiratory impacts of As would be nice.

Response: The unexpected finding of a significant relationship between arsenic and airflow obstruction in former smokers, not current smokers, puzzles our team. There is simply less known about nonmalignant lung disease outcomes and arsenic. A recent meta-analysis found the association between arsenic and FVC to be slightly stronger among non-smokers than smokers, also for reasons unknown.1 This finding, too, is surprising, as generally the quickest benefit after quitting cigarette smoking is improvement in lung function. This further points to the possibility that active smoking’s toxic effects could be masking those of arsenic.

Lung cancer studies which have examined the interaction between smoking, arsenic, and lung cancer, have generally found the presence of a synergistic interaction, that the excess risk resulting from the combination of exposure to smoking and arsenic is greater than the sum of excess risks from each exposure alone.2, 3 Mechanisms for synergism between arsenic exposure and smoking with lung cancer can include how cigarette smoke impedes tracheobronchial clearance, prolonging exposure of bronchial epithelium to arsenic-containing particulates, or arsenic deposited in the lung may enhance the multiplication of cells already promoting activities of additional carcinogens in tobacco smoke.3 Perhaps these mechanisms could help us investigate our own results with nonmalignant outcomes. But, this really is just conjecture for us at this stage. We have added the following to the paper:

A recent meta-analysis found the association between arsenic and FVC to be slightly stronger among non-smokers than smokers, also for reasons unknown.1 This finding, too, is surprising, as generally the quickest benefit after quitting cigarette smoking is improvement in lung function. This further points to the possibility that active smoking’s toxic effects could be masking those of arsenic; however, this is speculative.

Reviewer #2: GENERAL COMMENTS
Comment: Overall, this is a clearly written manuscript that reports associations between urinary arsenic concentrations and spirometric lung function in a large cohort of American Indians. The manuscript has a number of strengths, including the unique study population, sample size, use of study-specific spirometric reference values, careful statistical analysis, and adjustment for appropriate covariates, especially smoking. Despite these strengths, there are several limitations. Post-bronchodilator spirometry was not done, meaning that COPD cannot be properly assessed. Self-report of emphysema and chronic bronchitis is prone to outcome misclassification as the authors note. The relative inconsistency between the association of urinary arsenic concentration with odds of shortness of breath and that with the odds of having to stop because of shortness of breath is also problematic, suggesting potential participant misunderstanding of the symptom questions. In fact, the relative lack of association of most respiratory symptoms with urinary arsenic concentrations and the significant protective association with cough suggest that any effects of the low-moderate level arsenic exposure in this population are not of clinical importance. This interpretation is also supported by the small differences in lung function observed between 25th and 75th percentiles of urinary arsenic concentration. The Discussion section should be revised to include mention of the limited clinical significance of the findings.

Response: We agree that the post-bronchodilator spirometry was not done, however, this is typical of most population-based lung research, so we do not believe this is a flaw limitation for our study. This limitation, and others, are acknowledged in our discussion. Regarding the inconsistency between
shortness of breath and having to stop might be related to stopping while walking because of other issues (e.g. peripheral arterial disease, which was related to arsenic in this population). Because some of inconsistencies with the self-reported questions could be related to substantial measurement error, we prefer to focus our discussion on the objective spirometric measures. However, we agree that we need to be cautious in the interpretation of the findings and their clinical relevance. Additional research is needed to confirm whether our findings translate to clinical disease.

SPRCIFIC COMMENTS
Comments: Lines 11-112  As noted above, airway obstruction cannot be properly classified as COPD without post-bronchodilator spirometry. This should be acknowledged as a limitation in the Discussion.

Response: We have added this limitation to the discussion section: “We also could not confirm the presence of obstructive disease without post-bronchodilator spirometry.”

Comment: Line 149  "Mycobacterium" is one word.

Response: Mycobacterium is now written as one word in the manuscript.

Comment: Line 234  "People" or "individuals" would be better than "patients" here.

Response: “Patients” has been changed to “individuals.”

Comment: Line 235  Because no post-BD spirometry was done, "COPD" should be replace by "airflow obstruction."

Response: “COPD” has been changed to “airflow obstruction.”

Comment: Line 249  What is the basis for the assertion that "diabetes could be on the causal pathway between arsenic and restrictive lung pattern"? More explanation of the authors' reasoning would be helpful.

Response: In our study, diabetes was treated as a sensitivity analysis as the temporal and causal relationship between diabetes and lung function is debated. Cross-sectional studies have found that individuals with diabetes often have reduced lung function compared to those without diabetes.(4-6) but the definitive direction in addition to the pathophysiological mechanism to explain the association between lung function and diabetes is not known.(7) A cross-sectional study of older individuals without a history of chronic lung disease found duration of diabetes to be associated with reduced lung function, suggesting diabetes duration influences lung function more than glycemic control, and that obesity may also contribute.8 In a large longitudinal study (N= 27,711), a low FEV1 preceded and significantly predicted future diabetes, with the relationship not fully explained by inflammation, smoking, or obesity.9 There is likely a complex model of diabetes-related lung damage;(8) proposed mechanisms for the relationship between reduced lung function and diabetes include chronic inflammation, microangiopathy of lung vasculature, and loss of elastic recoil due to glycosylation of lung parenchyma.(9) With consideration to the body of research examining arsenic exposure and lung
function, previous studies have not adjusted for diabetes.(10) The SHS population has a high rate of diabetes, with prevalence close to 50%, and impaired lung function has been found to present before the development of diabetes.(12) Additionally, there is a large body of evidence suggesting that chronic arsenic exposure can contribute to diabetes development, further pointing to the need for a sensitivity analysis in our evaluation of the relationship between arsenic and lung health in the SHS. We have added the following to the discussion section:

Lung restriction in diabetes can result from chronic low-grade inflammation of the lung tissue; lung volume has been found to inversely correlate with the level of systemic inflammation, with a restrictive pattern of lung function loss associated with systemic inflammation.(14)

Comment: Lines 296-297 "select respiratory symptoms" is a bit misleading given the conflicting nature of some of the authors' results. Specifically listing "stopping for breath" as is done in the Conclusions of the Abstract would be better.

Response: We have added the specific symptom to the sentence, which now reads: “Our study provides evidence of an association between low-moderate arsenic exposure and a spirometric restrictive pattern, airflow obstruction (especially based on the LLN), and higher self-reported emphysema and stopping for breath.”

Works Cited


