Eboigbe et al. Response to reviewers:

We note our responses are in blue and we use the notations RxCx to define a specifically numbered comment (C) relating to a specifically numbered reviewer (R). RxARx refers to a specifically numbered Author Response (AR) that relates to a reviewer comment.

REVIEWER 1 (R1):

R1C1: The article "Mercury contamination in staple crops impacted by Artisanal Small-scale Gold Mining (ASGM): Stable Hg isotopes demonstrate dominance of atmospheric uptake pathway for Hg in crops" examines Hg in soil, crop, and atmosphere in the vicinity of ASGM operations. Authors measure THg and MeHg concentrations, as well as isotopic measurements of Hg. The paper is well written and data is clearly presented and discussed.

General comments:

The sampling design is well-structured, with multiple environmental sample types. The samples themselves are very valuable, as ASGM sites are understudied in the context of the global Hg cycle. The drawback of the study is that the number of samples per environmental sample type (soil, air, crop) is quite low, as the authors themselves state. Nonetheless, the authors used multiple analytical approaches to make the best use of these samples and the study is valuable for readers of Biogeosciences and researchers in the field, with minor corrections needed.

R1AR1: We appreciate the kind sentiments of the reviewer and also their understanding of the logistical, political, and ethical challenges of undertaking work of this nature. We appreciate the reviewer's understanding of the efforts made to get the most out of the samples that we were able to obtain from the mining and community partners.

Specific comments:

R1C2: Section 2.5.2. In the best practices for the analysis of Hg isotopes (e.g., as outlined in Blum et al., 2017: https://doi.org/10.2138/rmg.2017.82.17), the importance of using Tl internal standard is well explained. The authors do not report using Tl internal standard for their Hg isotope analysis. What is the reasoning behind not using it? In published syntheses of Hg isotopic work, studies conducted without the use of Tl internal standard are often excluded from data analysis.

R1AR2: We appreciate this comment and understand that it can raise concerns to some reviewers. From a strict scientific point of view, Blum and Johnson (2017) recommend use of Tl, but do not rigorously compare to the case of 'not using Tl'. From a more practical point of view, the Blum lab (and the reviewer's JSI lab) uses a Nu Instruments MC-ICPMS, which is an instrument known to be undergo large shifts in mass bias during a 24h session. Use of Tl helps correcting mass bias on the Nu and it is used in all Nu labs. We did our Hg isotope analyses in the Sonke lab on a Neptune MC-ICPMS, well known for its high stability of mass bias. Consequently, Tl is of little use on Neptune instruments. Note that both Blum and Sonke labs recommend sample matrix cleanup before analysis, which also helps avoiding matrix-induced mass bias and this was performed on all samples in this current study. Finally, on a Nu machine, the 12 Faraday cups allow measuring all Hg and Tl isotopes simultaneously. On many Neptunes, the 9 cups are physically restricted in their movement and do not allow collection of 203Tl, 204Hg, and 205Tl simultaneously; the Sonke lab therefore privileges measurement of 204Hg over Tl isotopes. To our knowledge, we are not aware of the exclusion of Sonke lab data, or other Neptune data (analysed without Tl) from review or synthesis works. The numerous high profile studies from both labs attest to the data quality; see for example Jiskra et al. (2021) which is published in Nature and uses the same methods without Tl-mass bias correction. Much of this discussion has been added directly to the supplementary information file.

Refs: Jiskra, M., Heimbürger-Boavida, L.E., Desgranges, M.M., Petrova, M.V., Dufour, A., Ferreira-Araujo, B., Masbou, J., Chmeleff, J., Thyssen, M., Point, D. and Sonke, J.E., 2021. Mercury stable isotopes constrain atmospheric sources to the ocean. *Nature*, *597*(7878), pp.678-682. https://doi.org/10.1038/s41586-021-03859-8

R1C3: Line 297: The d202Hg value of 0.29 ± 0.98 ‰ for Farm 1 does not indicate low variability as the authors state, ~1 ‰ SD is quite high for Hg isotopes. Please rephrase this discussion accordingly.

R1AR3: We will rephrase this sentence to the following:

"All soil samples exhibited **relatively small (compared to other contaminated soils)** variation in δ^{202}Hg (PS: 0.29 ± 0.98 ‰; Farm1 -0.26 ± 0.43 ‰) and Δ^{199}Hg MIF signal (PS: -0.09 ± 0.12 ‰; Farm 1 -0.07 ± 0.03 ‰) **(Grigg et al., 2018; McLagan et al., 2022; Vaňková et al., 2024)**."

Soils are a notoriously heterogeneous matrix. Thus, we do maintain that the changes, even the MDF, are small compared to what are found at contaminated sites (e.g., McLagan et al., 2022: δ^{202}Hg range ≈ 3-4 ‰; Grigg et al., 2018: range ≈ 1‰; Vankova et al., 2024: range ≈ 1.5‰) and these references have been added to the sentence.

Refs: McLagan, D.S., Schwab, L., Wiederhold, J.G., Chen, L., Pietrucha, J., Kraemer, S.M. and Biester, H., 2022. Demystifying mercury geochemistry in contaminated soil–groundwater systems with complementary mercury stable isotope, concentration, and speciation analyses. *Environmental Science: Processes & Impacts*, *24*(9), pp.1406-1429. https://doi.org/10.1039/D1EM00368B

Grigg, A.R., Kretzschmar, R., Gilli, R.S. and Wiederhold, J.G., 2018. Mercury isotope signatures of digests and sequential extracts from industrially contaminated soils and sediments. *Science of the Total Environment*, *636*, pp.1344-1354. https://doi.org/10.1016/j.scitotenv.2018.04.261

Vaňková, M., Vieira, A.M.D., Ettler, V., Vaněk, A., Trubač, J., Penížek, V. and Mihaljevič, M., 2024. Tracing anthropogenic mercury in soils from Fe–Hg mining/smelting area: Isotopic and speciation insights. *Chemosphere*, *357*, p.142038. https://doi.org/10.1016/j.chemosphere.2024.142038

R1C4: Lines 319-324: This paragraph seems out of place, fitting more to the introduction part into the justification for the chosen experimental design/methods (or somewhere else, but not here).

R1AR4: We believe this is the appropriate place in the manuscript because it provides direct comparison to another study of Hg in crops, specifically cassava, that examined inner peel, outer peel, and flesh. We use this study to highlight that this would be an advisable method of improving upon the work we have done (in future studies) to more conclusively identify if root epidermis/cortex does provide an effective barrier to Hg uptake and translocation to above ground tissue. We highlight the fit here by stating that the paragraph prior (lines 303-316) is specifically discussing the different uptake pathways (soil-to-root vs air-to-foliage).

R1C5: Lines 397-398: The subtraction of MDF for the soil-to-shallow roots (from Yuan et al 2022) is explained in supplementary material section S4. But until carefully reading the supplementary section, this subtraction is quite unclear to the reader, disturbing the reading flow. The authors should add a succinct explanation for this subtraction in the main text in lines 397-398.

R1AR5: This is a good suggestion, and we will make adjustments to more clearly define the two-end-member mixing model. Nonetheless, rather than adding this to the discussion, we deem it more appropriate to include this in the methods section (Section 2.6). Lines 227-229 will be updated to the following:

"A two-endmember mixing model was used to quantitatively determine source pathways for Hg in internal crop tissues according to Equation S4.1. The δ202Hg values of foliage for each crop are used as the first endmember: air-foliage uptake pathway. Similarly, the soil-root endmember must be the δ^{202}Hg signature of Hg immediately after uptake to the roots. Hence, the mean δ202Hg value for Farm1 soils minus the soil-to-shallow roots (roots above 150cm) MDF (ε^{202}Hg: -0.35) taken from Yuan et al. (2022) is used. Details of this two-endmember mixing model and the data used in the derivation of the endmembers are provided in Section S4."

R1C6: Lines 221-225, 392-395: The PTD analysis for Hg is not very robust. Therefore, authors should note that the conclusions they draw from these analyses are speculative (also in the discussion of the results, lines 392-395). Additionally, the term "speciation" is used too loosely, as speciation is, by definition, qualitative or quantitative measurement of chemical species, while in this case, there is no information about what the measured species are. Please replace "speciation" with "analysis" or "PTD analysis" in places where referred to PTD analysis, throughout the text.

R1AR6: We agree that PTD analyses are not highly robust, which is why we consider these analyses "quantitative and complementary" (lines 223-224). We also agree speciation is not the best terminology and we will change the sub-heading to "2.5.3 Hg speciation/fractionation analyses" and references to "speciation" to "speciation/fractionation" and "PTD speciation analyses" to "PTD analyses" as suggested.

R1C7: Lines 484-488: Would it make sense for future research to include some livestock near ASGM farms? Hg isotopic signatures in the livestock tissues could tell a very interesting story too. It could be worth mentioning in the text.

R1AR7: We concur will add the following statement at the end of the section the reviewer refers to:

"Adding Hg stable isotope analyses to any future work around Hg in livestock in ASGM areas could provide valuable insight into the biogeochemical processes involved."

R1C8: Section S3 and Figure S8: Authors mention certain reference standards were used for PTD analysis. Where can the reader see these desorption results for reference standards? The point of using these standards is to see if the desorption peaks of samples overlap with some desorption peaks of Hg standards. Why are the standards not shown then?

R1AR8: The standards we referred to here are from the three referenced studies [Biester and Scholz (1996), Mashyanov et al. (2017), and McLagan et al. (2022)]. We totally agree with the reviewer that when trying to identify specific fractions or species, it is best to cross reference to these standards. However, we stress that we deem these analyses "quantitative and complementary". The purpose of including the data presented in Section 8 (and referred to in lines 392-395) was to highlight the dual peaks observed in peanut and maize roots. This provides complementary data that support the notion of two Hg pools in the roots: one from direct uptake from soils (like in the epidermis/cortext) and another translocated to the roots from the air-foliage uptake pathway. These dual peaks that are clearly seen for peanut and maize roots were not observed in other plant tissues (Figure S8.1). We don't deem it necessary to compare to standards as these release peaks fall into the range of 190-300°C where many species/fractions are released (McLagan et al., 2022) and it would not be possible to accurately match these peaks to any specific species/fractions.

R1C8: Table S7.3: It would be clearer if authors wrote "Peanut soil 1, Peanut soil 2, ..." to make it clear that these are soil analyses and not crop analyses.

R1AR8: All samples in Table S7.3 will include soil in the sample labels in the revised version of the SI. The same will also be done for Table S7.2

Technical corrections:

R1C9: Line 31: "significantly high" should be "higher"

R1AR9: We will change this to "significantly higher" as this is a statistically tested difference.

R1C10: Line 40: "soil derived" should be "soil-derived"

R1AR10: This will be corrected.

REVIEWER 2 (R2):

R2C1: The manuscript describes a study on the distribution of Hg in soil, plants and the atmosphere near a ASGM mining site in Nigeria. The authors determined total and methyl Hg as well as stable Hg isotopes in soil, the atmosphere (by passive samplers (MerPas) as well as different parts of three types of edible plants with the aim to evaluate Hg levels in the plant and to track pathways of Hg uptake by the plants. Samples were taken near the mining site and at two farm sites (one reference site) situated at different distances from the mining site. The authors can show that the atmosphere, soil, and all edible plants are clearly affected by GEM emissions from the ASGM site. However, total and methyl Hg concentrations in all plant tissues were below reference dose thresholds.

Hg isotope analyses coupled with a two-endmember mixing model reveal that most Hg in plants are derived from atmospheric GEM uptake via foliage although crop roots appear to be to a larger extent influenced by Hg uptake from soil.

ASGM is seen as the most important todays anthropogenic Hg emission source to the environment. Besides its role in the global Hg cycle, investigations on the exposure of local people to ASGM Hg emissions, especially through crops, has been rarely investigated. Thus, the presented study is timely and important.

R2AR1: We thank the reviewer for their thorough summation of the study and positive feedback.

The study presents a comprehensive data set. I like such multi-proxy approaches as they offer deeper insights into the local biogeochemical cycling of Hg and disperion pathways. The manuscript is well written although I think that some parts esp. the abstract and the introduction could be shortened.

R2AR1: We appreciate the sentiments to reduce the length of the abstract and introduction with the aim of streamlining the manuscript. However, we are hesitant to do this as we believe we have worked hard to make these sections as concise as possible without removing context and background of the study.

We have already trimmed background from the abstract (384 words), and the remaining parts of the abstract list all the critical data and findings of the study. We believe any further cuts to the abstract will begin to reduce its impact and value.

With respect to the introduction (1086 words), it includes all the critical context of ASGM, Hg biogeochemical research into vegetation and crops, and Hg stable isotopes. Considering the breadth/interdisciplinarity of this work, we have to cover a broad background to give proper context for our work.

Furthermore, on June 9th we surveyed the abstract and introduction word counts of the 10 most recently papers published in *Biogeosciences*. The average word counts were 366±124 and 1213±221, respectively. We are close to the mean (within 1SD) of each; thus we do not deem the abstract or introduction to be of concerning length compared with other works in this journal. It is our preference to retain the abstract and introductions sections in their current form. At the advice of the Associate Editor we have converted the abstract into three separate paragraphs.

R2C2: The authors mentioned that Hg isotopes are used for both, evaluation of Hg species transformation processes and for tracking contamination or uptake pathways. In this sense, I believe that the interpretation of the Hg isotope data is probably not as robust as it seems. Especially because there is actually only a single Hg source which is GEM and Hg isotope fractionation proceeses in soils and plants are far from beeing completely known/understood. May be the authors could address this point in their discussion, although I don't think that this will change the overall message of the study.

R2AR2: We believe our assessment and treatment of both source and processing tracing is robust and both scenarios as essentially assessed in the 2-endmember mixing model. Since the burning of Hg-Au amalgams emits vast amounts of Hg(0) to air, we deem this the best estimate of our source. We have added the following statement to section 3.1:

"Considering the burning of Hg-gold amalgams emits Hg(0) directly into the atmosphere, we deem the mean stable isotope values for Hg(0) in air in the contaminated areas (PS and Farm1) to be signal most representative of the ASGM source."

We know stomatal assimilation imparts a big negative δ^{202}Hg fractionation and by comparing foliage δ^{202}Hg to air δ^{202}Hg (our "source signature") we have determined those fractionation factors for each plant (ε^{202}Hg values listed in the manuscript and abstract). We then use this value in foliage as our first endmember. Our second endmember becomes the value in soil adjusted for the sole study examining a δ^{202}Hg fractionation from soil to roots (as described in Section S4 and now in the updated Section 2.6 of the manuscript). We then consider any changes in δ^{202}Hg within the plant to be a result of mixing of these to uptake mechanisms. There could be some process-based fractionation occurring within the plant during translocation of Hg, but there has not been any studies that have identified any specific processes causing that or any fractionation factors associated with any of those potential processes. We cannot assume what we simply have no evidence for yet and we do not believe that we have made any such assumptions beyond what the literature has knowledge of (i.e., one study examining soil-to-root fractionation that is included in our analyses). We believe these two additions should suffice the request from the reviewer to more robustly describe source and process differences.

REVIEWER 3 (R3):

R3C1: The manuscript entitled "Mercury contamination in staple crops impacted by Artisanal Small-scale Gold Mining (ASGM): Stable Hg isotopes demonstrate dominance of atmospheric uptake pathway for Hg in crops" by Eboigbe et al. examines the biogeochemical cycling of mercury and uptake mechanisms in selected crops in areas contaminated with mercury due to ASGM. In light of the fact that ASGM represents the largest anthropogenic source of Hg today, and that due to the illegal nature of the ASGM activities, such areas are understudied, this contribution is very welcome and provides new valuable insights for scientific community.

Overall, the manuscript is very well and clearly written and logically structured. Appropriate complementary analytical methods are used, and the data are appropriately and adequately interpreted and evaluated in light of previous related studies. Therefore, I consider the article suitable for publication in the journal Biogeosciences. Regarding the content, I have only one general suggestion, namely that a section on the limitations of the studies be added to the discussion, where the aforementioned restrictions during sampling are critically evaluated and, on this basis, appropriate recommendations for future studies are made.

R3AR1: First we thank the reviewer for their very positive assessment of the manuscript. With respect to the limitations/recommendations, we have attempted to incorporate these into the text as each part of the study is address. However, we will add a Section 3.6 “Limitations and future work” to the manuscript that includes the following statement:

“As noted in Section 2.1, the scope of our sampling was limited by the social and geopolitical complexity of the ASGM issue. While it would have been optimal to assess larger crop sampling sizes at each site, we had to respect the wishes of the site operators, the community, and the farmers for whom these crops are their livelihood. Despite the lower-than-optimal sampling sizes, we achieved robustness through a thorough experimental design that captured samples from all the critical environmental compartments (and different plant tissues) and multi-method analyses. With that, we are confident in our data and the findings made with those data. Future studies should expand upon this work by adding dissected crop tissues (i.e., roots, edible parts) to improve the assessment of uptake pathways, internal cycling of Hg by plants, and translocation into edible tissues. Hg stable isotope analyses should remain a key part of future studies of this nature. Other studies have determined more elevated concentrations of Hg in edible parts of crops near ASGM areas (i.e., Adjorololo-Gasokpoh et al., 2012; Addai-Arhin et al., 2023); hence, if it is feasible, similar structured studies to our own should attempt to assess ASGM sites of differing (larger) scales and/or the proximity of farms to these sites.

This site was chosen due to existing partnerships that were built through discourse and trust. As described in Moreno-Brush et al. (2020), these partnerships between the communities, miners, local researchers, and international collaborators are critical to the success of Hg biogeochemical assays in ASGM areas. Security and research safety are considerable issues of research conducted in ASGM areas. While this should highlight the need for strong local partnerships, we stress that flexibility and adaption are vital components of such work, work which becomes increasingly important as ASGM continues to expand in the Global South.”

Here we stress the very real challenges of doing work in active ASGM areas. Many of these locations around the world are unsafe for researchers and are simply unfeasible. Therefore we highlight the importance of international collaborations and local partnerships in this section.