Responses to Referee 2 : MS No.: soil-2020-81

Title: Zinc lability and solubility in soils of Ethiopia – an isotopic dilution study

Author(s): Abdul W. Mossa et al.

Many thanks for your time and effort in reading and commenting on our manuscript; your detailed points will help improve it. In the following we address the individual points you raise.

Referee 2

General comments

The paper presents interesting research on zinc lability and solubility in Ethiopia.

Understanding the behaviour of zinc in these soils is essential in combating zinc deficiency, which is an issue affecting a quarter of the population in Sub-Saharan Africa. The authors explain in detail the influence of soil properties on different measures of zinc lability and show that these properties are more important than the total zinc content. The conclusion is that soil acidity is by far the most dominant factor, and for some soils the organic matter content plays a role. This information is useful in designing soil management strategies to improve zinc availability. The manuscript does not have a clear multidisciplinary context compared to other articles in SOIL, nevertheless the results may be relevant for a broad international audience.

Response:

We agree with the referee that the manuscript has a fairly constrained focus on technical aspects of Zn geochemistry in soils within the SSA region. Nevertheless, as such, the manuscript has important agronomical, health and economic implications given the importance of Zn deficiency in human populations—especially within SSA.

My overall impression is that this paper is very well written and contains a lot of relevant information. I have no comments on the introduction. A major shortcoming was found in the methods, where too little attention is paid to developing an adequate geochemical model.

Response:

Description of the use of the geochemical model will be improved in the revised manuscript as described below in our responses to specific comments from the referee below.

Next, I feel the results and discussion section is generally very interesting but could benefit from better organization. My main concern here is that relatively little attention is paid to the explanation of the study's own results, as the paper regularly reads like a literature review without clear reference to what those citations mean for the interpretation of data.

Response:

An important, and relatively novel, part of our manuscript is the isotopic dilution method that we used. Consequently, the focus of the manuscript is partly on method development and validation. Therefore, we have presented our results alongside the weight of evidence on the subject from the literature. This is particularly important because our data for the SSA soils shows a distinct (and perhaps unexpected) trend in E-value with pH compared with previous studies.

In addition, the introduction clearly states the problem of zinc deficiency, and as such the implications for soil management should be part of the discussion, instead of only a few sentences in the conclusion. Overall, this is a high-quality paper that with moderate revisions would be suitable for publication.

Response:

This is addressed below in the specific comments.

Specific comments

The title suggests an isotopic dilution study, while ID is only part of the work. The importance of the study seems to be to explain the effect of soil properties, for which ID was part of the methods but not the sole or main method. It is recommended to include 'soil properties' in the title, and remove 'an isotopic dilution study'.

Response:

Thank you for your suggestion. We believe that a substantial part of the novelty of the manuscript lies in the measurement of isotopically exchangeable Zn in soils from the study area, and the development and validation of the isotopic dilution method in this application. Therefore, we think that this should be emphasised and reflected in the title. However, we suggest a modified title including 'the effect of soil properties', as suggested.

Line 72: what determined the time of oven drying between 24-48 hours, and did that have an influence on the results?

Response:

The time required was determined by the texture and moisture content of the soils but in practice, this was a judgement, based on experience rather than prescription. For greater clarity, we suggest adding the phrase 'depending on the moisture contents of the soil samples' to the revised manuscript.

Please write in full the abbreviations the first time they are used (e.g. VWP, NPOC).

Response:

Thank you—we will define the abbreviations in full in the revised manuscript.

Some more attention should be given to the modelling using WHAM7, especially with respect to the models for Al, Fe and Mn oxides. What type of model is used and on which specific oxides is the parameterisation of the models based? What are the assumptions with respect to the specific surface area.

Response:

We suggest adding the following to the revised manuscript:

"The surface chemistry of oxides is simulated by a surface complexation model (Lofts and Tipping, 1998), which views the oxide surfaces as bearing hydroxyl groups that interact with protons and metal ions. Default values for specific surface areas in WHAM7 were used"

The input should be described more precise e.g. line 139 "Inputs to the model included cation and anion concentrations...." Specify which cations and anions and whether inputs are solution concentrations or concentrations in the soil solid phase.

Response:

Thank you for your suggestion—we suggest adding further details to the revised manuscript as follow:

"Inputs to the model included solution concentrations of cations (Na, Mg, Al, K, Ca, Mn, Fe, Co, Ni, Cu, Zn, Cd, Ba, Pb, U) and the anions ($NO3^{-}$, $PO4^{-3}$) in the solution phase of the $Ca(NO_3)_2$ soil suspensions"

Do the cations include Fe^{3+} and Al^{3+}? Tipping showed that these are important cations to consider because of their competition with other (trace) metals for binding to (dissolved) organic matter. In the case these cations were not measured their activities can be calculated from equilibrium with iron- and aluminium (hydr)oxide according to Tipping et al. 2002 (Geochimica et Cosmochimica Acta 66, 3211-3224)

Response:

Measured concentrations of Fe^{3+} and Al^{3+} in the solution phase of $Ca(NO_3)_2$ extraction were used as model input; this will be specified in the revised manuscript as described in response to your previous point.

A shortcoming of the study is the limited representativeness of the geochemical model for tropical (weathered) soils used to explain Zn lability and solubility. This stems from the assumptions made for the adsorptive constituents that are based on temperate soils, whereas the authors make it clear in the introduction that it is their ambition to study tropical soils, with the expectation that these will be different. This difference between tropical and temperate soils should then also be reflected in the model. The average fraction of humic substances was 36% for tropical soils modelled by Van Eynde et al. (2020): Boron speciation and extractability in temperate and tropical soils: A multi-surface modeling approach (Applied Geochemistry); this is notably different from the 50% used in the present study. In the study of van Eynde et al. it is also shown that the oxalate extractable Fe (non-crystalline oxides) is small compared to the dithionite extractable Fe (crystalline and non-crystalline) in such tropical soils. In the present study only oxalate extractable Fe is considered. Although the non-crystalline oxides have a much larger surface area than crystalline oxides, this may lead to an underestimation of the binding to iron oxides. Binding of metals to iron oxides is especially important at higher pH, which is the pH range for which the present study shows the highest overprediction of modelled soluble Zn.

Response:

The absence of characterization of soil organic matter (i.e. measurement of HA and FA) means that we are required to make certain assumption regarding the composition and reactivity of SOM. However, this is an approach which has been used frequently in previous studies and the 50% assumption that we make in the manuscript has been used extensively in the literature over a wide range of soils. The study you have cited may be qualitatively more relevant (HA 36% of SOM) but it reports the result for only 5 tropical soils. Therefore, we suggest retaining our initial approach but we have cited the study and noted the uncertainty associated with making such assumption in the revised manuscript. As we, and others, have noted in previous studies, the principal weakness of applying mechanistic geochemical assemblage models to whole soils (probably) lies in the crude nature of the measurements of geocolloidal adsorbents (oxides, HA, FA etc) and the assumptions regarding their characteristics (surface area etc) rather than in the model itself which has been parameterised using single, well characterised geocolloidal constituents.

In response, we suggest adding the following to a revised manuscript:

> "Van Eynde et al., (2020) found that the average fraction of humic substances in 5 tropical soils was 36%. However, the composition and the reactivity of soil organic carbon will be different from one soil to another. Consequently, there will be an associated error when relying on such assumptions in the absence of full characterization of soil organic carbon"

A positive point is the modelling of binding of Zn to Mn oxides which is usually not considered in multi-surface modelling studies for soils (see review Groenenberg and Lofts, 2014 Environ. Toxicol. Chem. 33, 2181–2196). The study shows that Zn binding to Mn-oxides may be highly relevant according to model predictions.

Response:

Noted, thank you.

Line 170: indicate what could cause the discrepancy between observed values and those reported for contaminated and uncontaminated soils.

Response:

The differences between the results for E-values reported in this manuscript and those reported in the literature are discussed in detail in section 3.2.2

Line 172: indicate why it is relevant that concentrations were positively skewed

Response:

Positive skewness indicates that the concentration of DTPA extractable Zn is low in most studied samples. This is indicated in lines 196-197

Figure 1: the added value of this figure in addition to Table 1 is unclear. Only two references are made to it (Line 172 and Line 183), and the skewedness of the data is not explained to be that relevant that it deserves a full figure. Additionally, inferences can be made about the skewedness by comparing the median and mean values in the min-max range as is done in Table 1. It is suggested to remove the figure.

Response:

Thank you for your suggestion, but we believe that readers will appreciate the nature of the dataset from a graph that encapsulates the distributions of the four indices of Zn bioavailability in soil, in their entirety. Therefore, we suggest it is better to keep the current figure.

Figure 2: it is unclear which label belongs to which line. PCA was also not explained in the statistics. It is felt that if the objective is to 'evaluate the correlation between soil variables' (Line 186) a correlation matrix is more intuitive than a PCA graph.

Response:

Thank your suggestion. We will improve the labels in the graph in a revised manuscript. We think that PCA graphs provide a more comprehensive and dynamic representation of the data than a correlation matrix and would therefore like to retain the PCA graph. However, we suggest also adding a correlation matrix to the supplementary material, as suggested.

Line 209: please add a brief final sentence on the overall method assessment and validation step

Response:

We suggest adding the following summary statement to the revised manuscript:

> 'Thus, given (i) the robustness of the ^{70}Zn distribution coefficient, (ii) the likelihood that the isotopic spike did not substantially affect the native Zn equilibrium or cause precipitation and (iii) the sub-micron filtration step and inter-laboratory agreement for Zn concentration in the 0.01 M Ca electrolyte soil suspensions, we are confident in the validity of the E-value determinations.

Line 220-223: unclear why this is relevant. In general, it is suggested to start with the most important explanations. It should be clearly explained why it is relevant to compare soils of the present study with urban or temperate agricultural soils when interpreting the pH effect.

Response:

Previous studies measuring isotopically exchangeable trace metals have been based on temperate soils (often contaminated) and urban soils. The weight of evidence from these studies suggests that %E-value (lability) declines with pH whereas the trend seen in our (uncontaminated) Ethiopian soils with their broad pH range differs from this. Therefore, it is important to discuss our results in the context of reported results in the literature and make comparisons that may potentially explain the difference between our results and those reported in previous studies.

Line 238-277: in the manuscript, this constitutes a one-page explanation of non-labile colloidal particles, leading to the conclusion that the correlation between ZnE and pH is genuine. This text can be shortened significantly. In addition, this part is more of a literature review, with relatively detailed accounts of the results found in other studies. What is missing is the link between the literature and the results found in the present study. For example, in line 266 the paragraph ends with the notion that solutions were filtered to 0.22 um; the authors fail to state what this means for their work and their data.

Response:

The study we cite (Tavakkoli et al., 2013) investigates the possible occurrence of non-isotopically exchangeable Zn in filtered soil suspensions. They found that the apparent proportion of isotopically exchangeable Zn was positively correlated with the pore size of the filter used (0.22, 0.45, 0.7 µm) - suggesting the presence of nano-particulate forms of Zn – whereas non-isotopically exchangeable Zn was absent when filtering to < 0.1 µm. As we used 0.22 µm filters in our study, we think that the non-isotopically exchangeable Zn in the filtered should be negligible. For greater clarity, the sentence "therefore, the existence of non-isotopically exchangeable Zn should be negligible." can be added to the revised manuscript.

Line 356-: In addition to the already mentioned possible explanations for overprediction also the presence of Zn-Al layered double hydroxides or Zn containing phyllosilicates could be considered (see already cited Bonten et al. 2008 and citations therein)

Response:

Thank you—we will cite this reference in the revised manuscript.

What is missing is a paragraph on the implications of the research for combating Zn deficiency. In the conclusion some 'tools' are mentioned, but this should be elaborated on at the end of the discussion. For example, it is concluded that it is soil properties rather than variation in total zinc that determines variability, but this is not translated in an overall conclusion on the conditions under which Zn deficiency occurs. A low solubility can still mean more Zn uptake if the total pool is larger, and vice versa.

Response:

This is a fair point. While our study does not specifically investigate crop uptake of Zn, we can address this in the Conclusions by re-working the text and adding an additional paragraph focussed on future work to link measurement of soil Zn availability with crop uptake**.** We suggest adding the following paragraph to the revised manuscript:

> 'Zinc uptake by crop plants will be determined by a combination of (i) soil Zn availability and (ii) plant controls over uptake and transport. It is likely that soil Zn availability will reflect both solubility and the magnitude of the reactive Zn reservoir that supports Zn^{2+} ion activity in the soil solution. For the Amhara soils studied it appears that (i) DTPA-extractable Zn provides the best estimate of the available Zn pool controlling Zn^{2+} solubility and (ii) that pH is the best predictor of the strength of labile Zn adsorption. Both assays are well recognised and fall within the scope of most agronomic laboratories. It therefore seems reasonable to suggest that Zn_{DTPA} and soil pH may, in combination, be tested as predictor variables for Zn uptake by staple crops in considering both crop Zn deficiency and biofortification for human health'

Technical corrections

Line 14: either explain what 'major knowledge gaps' are meant, or more generally indicate that SSA soil types are understudied.

Response:

Thank you for your suggestion. This can be changed to "many soil types in SSA are poorly studied" in the revised manuscript.

Line 76: comma or and after 'was determined'.

Response: Thank you—we will add a comma to the revised manuscript.

Line 135: remove comma after 'any deviation'.

Response:

Thank you—we will delete the comma and reorganise the sentence for improved clarity.

Line 176: significant but weak positive correlation

Response:

Thank you for your suggestion—we will add the phrase 'but weak' to the revised manuscript.

Line 190: explain what is meant with 'react in opposite ways.

Response:

For greater clarity, we suggest replacing the phrase with: "PCA analysis also shows that Zn_{Soln} and Zn_E appear to be inversely correlated, which is mainly a consequence of their opposite trends with pH" in the revised manuscript.

Line 206: 'but' = and

Response:

Thank you— we will replace 'but' with 'and' in the revised manuscript.

Line 224: 'changes' = differences

Response:

Thank you— we suggest replacing 'changes' by 'differences' in the revised manuscript.

Line 235: start a new paragraph on non-labile particulate matter.

Response:

Thank you for your suggestion, we suggest a new paragraph is started in the revised manuscript.

Line 273: unclear what is meant with 'magnitude of the trend'.

Response:

Thank you—for greater clarity we suggest the phrase is changed to 'strength of the trend'.

Line 282-283: unclear why this is important for the present study, should be explained.

Response:

It is vitally important to point out that results of the fractionation simulated by WHAM are for the reactive/labile fraction of Zn estimated by isotopic dilution method. This distribution does not cover the (pseudo-) total concentration as measured by aqua regia digestion; non-labile or 'fixed' metal in soil is not included in the adsorption equilibria described by geochemical models such as WHAM.

Line 302: the authors should make it clear that the pH-trend for Kdlab contrasts the one found for ZnE, instead of leaving it for the reader to infer.

Response:

Thank you for your suggestion. The trend of Zn_E with pH is discussed elsewhere (3.2.2). Although, Kd is one of two components in calculating Zn_E, they are different/separate. Therefore, we think there will be no added value to make such a contrast.

Line 316: the 'p' looks like rho.

Response:

Thank you for spotting that, this will be corrected to 'p' in the revised manuscript.

Line 316: 'some influence on metal adsorption strength would be expected', this is vague and should be explained more clearly. In general, the remaining sentences of this paragraph should be elaborated on, to further clarify the pH-effect, and contain a brief conclusion.

Response:

We simply mean that an effect of soil organic matter on metal solubility would be expected because of its importance as a metal adsorbent. However, the nature of the effect is complicated by the opposing effects of organic matter in the solid and solution phases – as discussed. In the revised manuscript we suggest changing the text to:

> "some influence on metal adsorption strength would be expected because of the importance of humus as a metal adsorbent (Fan et al., 2016)."

The rest of the paragraph then explains the two opposing effects of organic matter in the solid and solution phases, which may account for the relatively weak overall effect on metal solubility.

Line 323: comma after 'strength of adsorption'

Response:

Thank you—we will add a comma to the revised manuscript

Line 361-365: This can be shortened, as all cited studies conclude the same.

Response:

Thank you for your suggestion. Even though the two references are similar we think they provide a slightly different explanation (e.g., specifying calcite and hydroxyapatite as binding phases). Therefore, we suggest it is better to keep them separate.