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Peer Review File

In-situ synthesis of iron-based reactive nanoparticles for groundwater remediation in heterogeneous two-and-three-dimensional porous media

Corresponding Author: Dr Carlo Bianco

This manuscript has been previously reviewed at another journal. This document only contains information relating to versions considered at Communications Earth & Environment.

This file contains all editorial decision letters in order by version, followed by all author rebuttals in order by version.

Attachments originally included by the reviewers as part of their assessment can be found at the end of this file.

Version 0:

Decision Letter:

** Please ensure you delete the link to your author home page in this e-mail if you wish to forward it to your coauthors **

Dear Dr Bianco,

Your manuscript titled "In-situ synthesis of iron-based reactive nanoparticles for groundwater remediation: controlled emplacement in 3D and heterogeneous 2D porous media" has now been seen by 2 reviewers, whose comments are appended below. You will see that they find your work of some potential interest. However, they have raised substantial concerns that must be addressed. In light of these comments, extensive revisions will be required before we can further consider the manuscript for publication. We would, however, be interested in considering a revised version that fully addresses these serious concerns.

We hope you will find the reviewers' comments useful as you decide how to proceed. If additional work allows you to either incorporate or refute these criticisms, we will be happy to look at a substantially revised manuscript. If you choose to take up this option, please either highlight all changes in the manuscript text file, or provide a list of the changes to the manuscript with your responses to the reviewers.

To be publishable in Communications Earth and Environment, the following editorial thresholds need to be met

**Demonstrate with analysis and references whether sulfur-based reductants can reduce Fe(II)/Fe(III) to Fe^0 and clarify possible Fe–S formations.
**Provide stronger and more convincing proof of Fe^0 presence, supported by quantitative yield assessment and clear differentiation from oxidized or sulfide phases.
**Clearly describe the composition, concentration, and injection protocol of the “iron liquid,” ensuring reproducibility for 3D and 2D experiments.
**Clarify that degradation tests were batch-based, justify concentrations, explain why flow tests weren’t done, and distinguish Fe^0 effects from direct reductant reactions.

When resubmitting, please provide a point-by-point response to the reviewers’ comments. Please submit your responses as a separate file, distinct from your cover letter where you can add responses to the Editors’ comments that you do not want to be made available to the reviewers. Word files are preferred. We recommend that any figures, tables or graphs that are included in the response to reviewers are also included in the main article or Supplementary Information.

Please bear in mind that we will be reluctant to approach the reviewers again in the absence of substantial revisions.

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elsewhere in the meantime.

We are committed to providing a fair and constructive peer-review process. Please do not hesitate to contact us if you wish to discuss the revision in more detail.

Please use the following link to submit your revised manuscript, point-by-point response to the reviewers' comments with a list of your changes to the manuscript text (which should be in a separate document to any cover letter), a tracked-changes version of the manuscript (as a PDF file) and any completed checklist:

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Please do not hesitate to contact us if you have any questions or would like to discuss the required revisions further. Thank you for the opportunity to review your work.

Best regards,

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REVIEWER COMMENTS:

Reviewer #1 (Remarks to the Author):

Manuscript#: COMMSENV-25-4663-T
Title: In-situ synthesis of iron-based reactive nanoparticles for groundwater remediation: controlled emplacement in 3D and heterogeneous 2D porous media
Corresponding Author: Carlo Bianco

The manuscript titled "In-situ synthesis of iron-based reactive nanoparticles for groundwater remediation: controlled emplacement in 3D and heterogeneous 2D porous media" presents a novel strategy for the in-situ generation and emplacement of reactive nanoscale zero-valent iron (nZVI) within contaminated aquifers. The authors propose the co-injection of environmentally benign precursor solutions, leading to the formation of reactive iron nanoparticles directly inside the porous medium. The approach is evaluated through a combination of 3D homogeneous and 2D heterogeneous laboratory-scale experiments, which demonstrate improved particle distribution and larger reactive zones.

Overall, the study addresses a relevant and long-standing challenge in the field of subsurface remediation—namely, the limited mobility and uneven distribution of injected nZVI in heterogeneous aquifers. The concept of producing reactive

particles in situ through precursor reactions is indeed innovative and potentially impactful for large-scale applications. However, several fundamental scientific issues related to the reaction chemistry, product identification, and mechanistic understanding remain unresolved.

1. The central assumption of this work—that sulfur-based reductants such as sulfites, bisulfites, thiosulfates, dithionites, or related compounds can reduce Fe(II)/Fe(III) salts to zero-valent iron (Fe^0) under aqueous conditions—is thermodynamically questionable. The standard reduction potentials of these sulfur species suggest that they are capable of reducing Fe(III) to Fe(II), but not Fe(II) to Fe0. Under the experiment conditions of manuscript, the likely product nanoparticles are Fe(II) species or Fe–S minerals (e.g., FeS, FeS2, Fe3S4) rather than Fe0. The manuscript should critically evaluate this aspect with thermodynamic analysis and literature references.

2. The analytical evidence presented in the manuscript does not convincingly demonstrate the formation of zero-valent iron. SEM–EDS provides only elemental composition and cannot distinguish Fe0 from oxidized or sulfide phases. The reported XPS data are also insufficiently detailed to substantiate the claimed Fe0 species. The authors are strongly encouraged to provide more definitive characterization using XRD, high-resolution TEM, and Mössbauer spectroscopy, along with appropriate control samples.

3. The reported Fe0 production yields (60–94%) appear remarkably high, yet the basis of these values is not clearly explained. How much of the total Fe in the 3D and 2D experiments is truly Fe0? A quantitative determination—such as acid digestion followed by H2 quantification—is needed to support this claim. Without such evidence, the interpretation of Fe0 formation remains speculative.

4. The proposed mechanism implies a two-step electron transfer process: (i) electrons from sulfur-based reductants first reduce Fe ions to Fe^0, and (ii) the generated Fe^0 subsequently transfers electrons to chlorinated solvents (TCE, PCE) for dechlorination. This indirect route could lead to lower overall electron utilization efficiency, compared with direct reductive degradation by dithionite itself. Moreover, dithionite and related species are known to reduce TCE directly under certain conditions. The authors should discuss whether their approach offers any net advantage in electron efficiency and whether the observed dechlorination could partially arise from the reductant rather than from Fe0.

5. In realistic aquifers, abundant natural oxidants (e.g., dissolved oxygen, nitrate, Mn(IV), Fe(III) oxides) would rapidly consume sulfur-based reductants before they can react with Fe salts. The authors should discuss how their proposed method would perform in such complex chemical environments.

Reviewer #2 (Remarks to the Author):

Please see attached.

Version 1:

Decision Letter:

Dear Dr Bianco,

Your manuscript titled "In-situ synthesis of iron-based reactive nanoparticles for groundwater remediation: controlled emplacement in 3D and heterogeneous 2D porous media" has now been seen by our reviewers, whose comments appear below. In light of their advice we are delighted to say that we are happy, in principle, to publish a suitably revised version in Communications Earth & Environment.

We therefore invite you to revise your paper one last time to address the remaining concerns of our reviewers. At the same time we ask that you edit your manuscript to comply with our format requirements and to maximise the accessibility and therefore the impact of your work.

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We hope to hear from you within two weeks; please let us know if you need more time.

Best regards,

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REVIEWERS' COMMENTS:

Reviewer #1 (Remarks to the Author):

The authors have made substantial efforts to address the previous comments, and their point-by-point responses have addressed most of major concerns. I only suggest the following minor revisions before further consideration or acceptance:
1. The Graphical Abstract should be revised to better reflect the current manuscript. In particular, the presentation of nZVI

and Fe0 is not fully consistent with the revised text. Since the manuscript has replaced nZVI with nRIM, the Graphical Abstract should be updated accordingly.
2. Please standardize the terminology throughout the manuscript. At present, both “nZVI-based particles” and “nanoscopic Reactive Iron Minerals” are used. The authors should clearly define which term is preferred and use it consistently throughout the text, figures, and supplementary materials. In my view, “nZVI-based particles” may not be the most appropriate term, because the current evidence does not allow the authors to determine how much nZVI is actually present in the material.
3. The manuscript would benefit from clarification of the degradation products of PCE and TCE by nRIM. This information may help elucidate the dechlorination pathway and provide useful clues regarding the reactive components and degradation mechanism of nRIM.

Reviewer #2 (Remarks to the Author):

Authors were able to address all comments to improve the manuscript, thank you. No further suggestions.

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Manuscript COMMSENV-25-4663-T

"In-situ synthesis of iron-based reactive nanoparticles for groundwater remediation: controlled emplacement in 3D and heterogeneous 2D porous media"

by Andrea Gallo, Leonardo Magherini, Federico Mondino, Carlo Bianco*, Tiziana Tosco, Rajandrea Sethi

In this document we present our point-by-point reply to the reviewers' comments. The comments are reported in in regular typeface (blue), whereas our responses are written in regular typeface (black). Line numbers refer to the revised version of the manuscript.

RESPONSE TO REVIEWERS

We sincerely thank the reviewers for carefully reading the work and for their constructive criticisms and valuable comments. Below, you will find a point-by-point description of each question and how each comment was addressed in the manuscript. The original reviewer's comments are reported in regular typeface (blue), our responses in regular typeface (black), and the modified manuscript text in red. Line numbers refer to the revised version of the manuscript.

REVIEWER #1

1. The central assumption of this work—that sulfur-based reductants such as sulfites, bisulfites, thiosulfates, dithionites, or related compounds can reduce Fe(II)/Fe(III) salts to zero-valent iron (Fe^0) under aqueous conditions—is thermodynamically questionable. The standard reduction potentials of these sulfur species suggest that they are capable of reducing Fe(III) to Fe(II), but not Fe(II) to Fe0. Under the experiment conditions of manuscript, the likely product nanoparticles are Fe(II) species or Fe–S minerals (e.g., FeS, FeS2, Fe3S4) rather than Fe0. The manuscript should critically evaluate this aspect with thermodynamic analysis and literature references.

   Response: we thank the reviewer for the insightful comment. We concur that sulfur-based reductants are not as strong as borohydride, which is usually the preferred reductant for the wet-chemistry synthesis of nZVI. Literature works show that batch synthesis with sulfur-based reductants such as dithionite lead to formation of Fe(0), albeit with lower yields and poorer remediation performances than what can be obtained with borohydride (Ma et al., 2016; Sun et al., 2011). However, considering dithionites, the low reduction potential is only reflected by the direct reduction from dithionite itself. There is a second possible reduction pathway discussed in literature, based on the decomposition of dithionite in the sulfur dioxide radical (also called dithionite radical) $SO_2^{-\bullet}$ (Burlamacchi et al., 1969; Mayhew, 1978; Neta et al.,

1988). In these studies, the redox potential was found to be strongly pH-dependent and while at acidic pH it shows value insufficient to reduce Fe(II) to Fe(0), the redox potential is reported -0.66 V at pH 7 and as low as -1.13 V at pH 14 (Burlamacchi et al., 1969; Mayhew, 1978). With such values, the reductant solution has the required properties to effectively reduce Fe(II) to Fe(0) and the reagent formulation applied in this work purposedly exploits an alkaline pH to enhance the reduction potential of sulfur-based reductants. This was briefly mentioned in the original manuscript in lines 99-101 "that under specific conditions showed sufficient reduction potential to promote the formation of ZVI". We have also expanded the relevant literature references to the manuscript (line 101-102) to substantiate the choice of reducing agent, but we believe an in-depth thermodynamic analysis is beyond the scope of this paper.

We also agree that formation of other byproducts, especially iron sulfides and oxides, is a possibility as also been reported by previous works (Fan et al., 2017; Ma et al., 2016; Sun et al., 2011). For example, we cannot exclude that some of the oxides detected in the XPS analysis were already formed in-situ, in addition to their formation from the oxidation of the nZVI after the synthesis. Nonetheless, many of the possible other iron-based products have also shown interesting properties for remediation, for example iron sulfides, iron oxides and green rust (Butler & Hayes, 1998; He et al., 2015; O'Loughlin & Burris, 2004). To reflect this possibility of a mixed composition, and to also answer the following point raised by the reviewer, the entire paper was revised exchanging the term "nZVI" with "nanoscopic Reactive Iron Minerals" (or nRIM) or "ZVI-based nanoparticles" when referring to the materials synthetized with the proposed protocol.

2. The analytical evidence presented in the manuscript does not convincingly demonstrate the formation of zero-valent iron. SEM–EDS provides only elemental composition and cannot distinguish Fe0 from oxidized or sulfide phases. The reported XPS data are also insufficiently detailed to substantiate the claimed Fe0 species. The authors are strongly encouraged to provide more definitive characterization using XRD, high-resolution TEM, and Mössbauer spectroscopy, along with appropriate control samples.

Response: we agree with the reviewer that Mössbauer spectroscopy is the best technique to assess iron speciation. As we do not have such instruments available in our facilities, we actually attempted to send samples collected from the 3D experiment to an external laboratory for Mössbauer analysis. However, despite the precautions taken all the samples underwent oxidation during transport and storage in the facility before analysis (with visual evidence of the material having turned red from the initial black color) and therefore we were unable to obtain usable data. The high reactivity of the material when exposed to air proved very challenging for the material characterization. The analysis of the in-situ synthetized material requires the desaturation of the tank, excavation of the sand where synthesis/deposition occurred and lastly separation of the particle not adhered to the sand. Each of these steps involves exposure to air, and it was possible to visually observe the color of

the particle shifting from gray/black to brown/red. We opted for the XPS analysis as the instrumentation was available in our building, allowing us to reduce the time the sample was exposed to air, or stored before measure. We added the two Ar sputtering step to explore the depth profile of the nanoparticles, removing the oxide layers that could have formed during handling. The data revealed that there is indeed a zero-valent iron core in the particles, and that the oxide layer is compatible with the oxidation of a ZVI material. Nonetheless, we do agree that the presence of other reactive iron phases cannot be excluded and we changed the terminology for our material to "nanoscopic Reactive Iron Minerals" or "ZVI-based nanoparticles".
We agree with the reviewer that SEM-EDS analysis is not sufficient to prove the presence of zero-valent iron, but its purpose in the current work is to show that a portion of the particles strongly adheres to the sand grains, thus forming a non-mobile long-term reactive zone in the aquifer.

3. The reported Fe0 production yields (60–94%) appear remarkably high, yet the basis of these values is not clearly explained. How much of the total Fe in the 3D and 2D experiments is truly Fe0? A quantitative determination—such as acid digestion followed by H2 quantification—is needed to support this claim. Without such evidence, the interpretation of Fe0 formation remains speculative.

   Response: we thank the reviewer for the comment, and we have revised the entire section to address this issue. The data presented on the product yield were not intended to convey the yield in terms of Fe(0), rather in terms of total iron deposited or iron conversion into mineral form. This is to address the efficiency in using the iron reagent and also to assess how much free iron could affect downstream receptors. We have revised the entire section (lines 270, 405, 414-415) specifying that the yield refers to the total deposed iron (obtained from the ICP-MS data) and not to actual Fe(0) production. We also updated the terminology for the nanoparticles (from "nZVI" to "nanoscopic Reactive Iron Minerals" or "nZVI-based") to better convey this message.
   Acid digestion for hydrogen evolution was attempted, and we were able to observe bubbling from the material. However, no reliable data could be obtained due to the low amount of reactive material available and the lack of instrumentation suited to measure the small quantities of H_2 therefore produced.

4. The proposed mechanism implies a two-step electron transfer process: (i) electrons from sulfur-based reductants first reduce Fe ions to Fe^0, and (ii) the generated Fe^0 subsequently transfers electrons to chlorinated solvents (TCE, PCE) for dechlorination. This indirect route could lead to lower overall electron utilization efficiency, compared with direct reductive degradation by dithionite itself. Moreover, dithionite and related species are known to reduce TCE directly under certain conditions. The authors should discuss whether their approach offers any net advantage in electron efficiency and whether the observed dechlorination could partially arise from the reductant rather than from Fe0.

Response: we understand the point raised by the reviewer, that the complex Fe(0)/Fe(II)/Fe(III) chemistry may reduce the efficiency of the electrons provided by dithionite, which can be effective for the remediation of some compounds. However, dithionite is able to induce degradation of some contaminants only, and in most applications it is coupled with engineered nanomaterials (Nunez Garcia et al., 2016; Xie & Cwiertny, 2010), natural occurring minerals (Amonette et al., 1994; Song et al., 2019) or other chemical agents (i.e., persulfate) (Brown, 2010; Song et al., 2019) as its efficiency is strongly related to pH and decades rapidly. Moreover, the use of dithionite in ISCR-like processes does not ensure sufficient groundwater persistence to achieve effective aquifer remediation. In fact, dithionite, being a liquid reagent, may be quickly washed away by the natural groundwater flow, and successive re-injection are often needed to accomplish satisfactory contaminant removal. This is particularly true in the presence of medium-low permeability lenses that, after the successful remediation of the more permeable portion of the aquifer, will act as secondary sources releasing the contaminant by backdiffusion (Kueper et al., 2014). The main advantage of a remediation approach based on the formation of Reactive Iron Minerals, among which nZVI-based material, is the formation of a stable reactive zone composed of solid reactive particles which can exert a reductive activity for longer times, and has also the potential to be restored with follow-up reductive agents injections (Xie & Cwiertny, 2010).

To minimize the possibility of a combined effect of nanoparticles and residual reductant on the PCE/TCE degradation, the reactive material was thoroughly washed before its use in the batch tests. We expanded this section of the manuscript accordingly in lines 296-298:

"To minimize a possible contribution to the reduction by residual reducing agent, the 3D tank was flushed with 5 volumes of tap water between the end of the synthesis and the excavation protocol used to collect the material."

And lines 302-303:

"This particles were washed using a volume of degassed deionized water equal to 5 times the supernatant volume at the end of the synthesis, divided in 5 aliquots".

For the particles extracted from the 3D experiment, the degradation shown is already corrected from the losses observed using a control sample of the sand collected downstream the injection well but in an area without any deposed material (even so, such losses were below 1%). We expanded line 295 to add this detail in the manuscript.

5. In realistic aquifers, abundant natural oxidants (e.g., dissolved oxygen, nitrate, Mn(IV), Fe(III) oxides) would rapidly consume sulfur-based reductants before they can react with Fe salts. The authors should discuss how their proposed method would perform in such complex chemical environments.

   Response: we agree with the reviewer that the conditions explored in the present work are ideal and far from a real aquifer. We tried to bridge the gap regarding the groundwater using tap water, instead of deionized water, to saturate the porous medium, to create the background flow and to prepare all the reagents injected (line 130). Regarding the porous media, it is true that real materials would contain mineral phases and/or organic matter which would contribute to the consumption of the reductant.
   However, the same situation is already encountered in in-situ chemical remediation (both ISCO and ISCR), for which the site natural demand for the amendment (either oxidant or reducing agent) is accounted with an overdosage of the same. This same principle is already applied in the protocol presented, as the reducing agent is injected first to condition the aquifer redox potential to a range suitable for the synthesis of Fe(0), and to account for the possible higher oxygen content due to the packing of the porous medium. Therefore, either a longer first injection or a more concentrated solution can be used in real applications to initially deplete the natural oxidants before starting the injection sequence for the actual synthesis.

REVIEWER #2

1. The abstract is misleading in lines 34-35. Authors performed batch experiments with nZVI synthesized in the 3D aquifer. This sentence makes the reader believe that chloroethenes (PCE and TCE) were removed within 21 days in-situ, suggesting the technology could effectively be used for nanoremediation. Authors tested that the nZVI produced in the 3D aquifer can remove these compounds in batch experiments only (discussed in sections 2.3.4 and SM6). Authors did not attempt to inject chloroethenes into the 3D or 2D aquifers, therefore the reader is left without an explanation on why the reactivity of the nZVI produced in the aquifer was not tested under flow conditions. Proving that nZVI can be synthesized and effectively reduce a groundwater plume (whether in the 3D or 2D setup) would have been an important contribution. Authors should clarify why treating a contaminant plume in-situ was therefore not tested. This was only mentioned at the end of the manuscript (after lines 512) as a perspective. The 3D model aquifer has various piezometers that could have been used to track the contaminant plume and validate the approach. Geochemical interpretation as in Figure SM8 would have been possible.

   Response: we thank the reviewer for this comment, we have modified the sentence specifying "batch degradation of chlorinated ethenes" in line 34-35. We recognize the importance of assessing the reactivity of the synthetized materials under flow-conditions in order to bring the proposed protocol closer to readiness for a pilot test, and treating a contaminant plume in the same 2D/3D tanks after synthesis would have been indeed of great interest. At the present stage, we opted for batch tests due to the high complexity involved in our 3D/2D set-ups. Especially for the 3D set-up, very large handling volumes for the contaminated solution are required, and we do not have the facilities to conduct experiments of this scale with volatile toxic compounds abiding safety requirements. Secondly, many components of the 2D/3D tanks are non-inert plastic (acrylic, PVC) which would cause significant absorption of the chlorinated ethenes masking the degradation operated by the emplaced materials. Tests in flow conditions would also require a control (or blank) test performed to evaluate volatilization adding to the complexity and the use of toxic chemicals. Given the high uncertainty we predicted on the final data, we preferred to wait to perform reactive tests under flow conditions for a later time opting to prove the potential remediation activity of the in-situ formed material with batch tests. We revised the manuscript in lines 283-285 to substantiate our choice for batch experiments: "Degradation tests under flow conditions in the 3D and 2D tanks were deemed too complex for the present work, due to the adsorption of contaminants on the plastic components (piezometers, tank body), volatilization and for safety concerns in the handling of large volumes of toxic chemicals.".

2. The batch experiment showed that the removal efficiency of the produced nZVI in the 3D aquifer was between 30 and 40% for PCE and TCE, respectively (lines 441-443 and SM7). This removal efficiency is low. Why did authors choose a parallel test with nZVI produced with a batch synthesis protocol at 1 g/L and not at 0.1 g/L

as formed in the 3D aquifers (Figure SM7)? What is the reasoning of testing concentrations at 20 and 10 ppm of TCE and PCE, respectively?

Response: the low removal efficiency is mainly caused by the severe oxidation that the material was subject to during the excavation procedure to expose the different layers used in the tomographic reconstruction. We were also limited in the amount of material that we could use as from a single 3D experiment we performed total iron measurements with ICP analysis following ISO methods, which required a significant amount of material. Therefore, the tests at 0.1 g/L are under a condition where the reactive material is the limiting reagent, as emerges from the curves approaching a plateau at 21 days. In addition, when normalized for the reactive material dose the reaction rates observed are not too dissimilar from what reported in the literature (discussion in lines 462-469). The test performed at 1 g/L with a batch protocol aims to display the degradation ability of the same material when subject to minimal oxidation at a dosage more consistent with a possible future implementation. In this way we aimed to overcome the aforementioned limitations linked to using the material from the tank experiments. The PCE and TCE concentration were selected based on our previous experience characterizing water samples from real contaminated sites, to provide a plausible scenario to test the material performances. We added this detail in lines 288-290 "The concentrations for these contaminants were selected based on our group previous experience with groundwater samples from contaminated sites in Italy.".

3. Authors validated the synthesis of the nZVI in a 3D medium-scale homogeneous aquifer using a tomography-like approach, which showed the production of nZVI around the injection well. A mixture of ferrous chloride, tap or deionized water and an iron liquid solution was induced. Authors mentioned (lines 127-133) that the in-situ iron liquid consists of a blend of various reducing agents, including sulfates, sulfites, bisulfites, metabisulfites, dithionites, dithionates, thiosulfates, and pH modifiers. However, the reader is left without a proper understanding of how this iron liquid solution works, what the ratios and quantities in this solution are (how much is injected?), and so how this can be reproducible. One wonders: why not provide the exact composition of the solution? Authors provided the chemical characterization of the tap water (SM1), the injected concentration of the ferrous chloride but not much information on this iron liquid solution which is key for the production of nZVI. Is the solution commercially available? If not, it is not indicated in the manuscript if there is a confidentiality about the iron liquid solution. Could authors be more explicit about the composition of this solution and the concentrations injected? This would be of interest to the remediation industry who could test this approach and make the proper calculations (stoichiometry) for the synthesis of nZVI and projected contaminant removal. Later in the text (Table 1) we read about the duration of the injection steps and cycles for solutions R, TW (water) and S in the 3D aquifer, but without the concentration of solution R (the iron liquid). We read again something similar in section 2.4.3 for the 2D aquifers. In line 340, authors mentioned that their protocol resulted in the production of 160 mg of iron

(assuming a yield of 100%) in the 2D aquifer. This iron liquid solution, composition, concentrations and the exact protocol of its injection should be clarified. This is necessary for scale up and for testing in other aquifer setups, pilot tests and real scenarios.

Response: we thank the reviewer for the insightful comment. As he/she correctly hypothesizes, ISI-Liquid ("In Situ Iron Liquid") is a commercial product and part of a patented and proprietary technology, as disclosed in our submission, and we are not authorized to fully disclose its exact composition. However, the product is commercially available and can be purchased from any researcher or industry for testing. Also, we did not realize that the dosage of ISI-Liquid used in the study was missing, and we thank the reviewer for raising this point. Following the reviewer's comments, we have added details on both the reagent concentration and the injection protocol adopted (lines 132-136), thus ensuring reproducibility of the 3D and 2D experiments after purchase of the commercial formulation ISI-Liquid from the official supplier.

4. MINOR COMMENTS

a. In the abstract: Please consider removing environmentally relevant contaminants and mention chloroethenes.

   Response: we have modified the abstract (line 35) accordingly

b. Line 75: Please clarify the tendency of contaminants to accumulate in low-permeability layers. Contaminants are affected by different processes. Is it because of sorption, diffusion, density (DNAPL) or due to low-hydraulic conductivity? Is this particularly important for chloroethenes?

   Response: we have modified the manuscript accordingly in lines 75-79 "It is well known that diffusion leads organic contaminants to accumulate in low permeability layers, from which they are subsequently slowly released by back-diffusion phenomena. In such cases, after conventional treatment (advective-driven) the low hydraulic conductivity leads to an incomplete remediation, and these lenses act as secondary sources"

c. Line 99: Please clarify which natural reducing agents. This would help linking geochemical conditions in natural aquifers.

   Response: we have specified the most commonly tested natural reductants and provided the literature references in line 103 "such as polyphenols, vitamins, flavonoids".

d. Line 175. The hydrodynamic parameters of the porous medium were obtained from column tracer tests performed in the same material .... I think it should be read with the same material. Tracer tests were not tested in the 3D or 2D setup. Authors should provide information on why tracer tests were not performed in the aquifers.

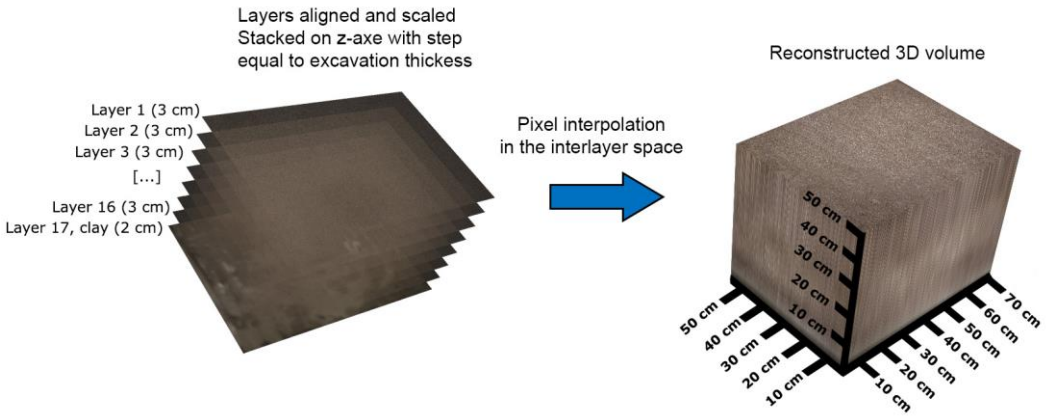
Response: we have modified “in” with the suggested “with”. Additionally, following the reviewer’s suggestion we have specified in line 182-183 of the manuscript that the hydrodynamic parameters were also validated with a tracer test performed in the 3D tank “The obtained parameters were validated in the 3D tanks with a tracer test prior the synthesis, the results are briefly presented in Supplementary Materials.”. Additional details are presented in Supplementary Materials.
The choice to use column test as primary method is for the higher accuracy compared to higher uncertainties deriving from the piezometer and sampling system in the 3D tank.

e. Line 238: When was the excavation process performed for the 3D aquifer and the tomography-like approach? Please clarify how many pore volumes passed so the reader can have an idea how long it took to produce the nZVI. Was the aquifer drained prior to the excavation?

Response: the excavation was performed 10 hours after the synthesis, during this time (overnight) a total of 5 pore volumes of water flowed through the tank. This step is not part of the synthesis and was only performed to wash out the reductant residues and verify the stability of the emplaced nanoparticles. For the injection, the total duration (which consists of the times reported in Table 1 plus the 20 min tap water steps between each cycle mentioned in line 230-240) is approximately 124 minutes of total injection, not considering the manual time to switch pumps.
During the excavation we opted for a progressive desaturation of the tank, to prevent as much as possible the exposure to air of our reactive material. We desaturated approximately 4 cm at the time, while excavating layers of 3 cm. We have added this detail to the Supplementary Materials.

f. Figure SM3: I recommend adding the distances x, y as similarly presented in Figure 3B.

Response: we have modified the image accordingly.



g. Line 396: The deposition yield wouldn't be between 60 and 94% for external and inner areas of the reactive zone, respectively? Please check this sentence.

Response: we thank the reviewer for the comment. This was indeed a typo, and we have corrected the manuscript.

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"In-situ synthesis of iron-based reactive nanoparticles for groundwater remediation: controlled emplacement in 3D and heterogeneous 2D porous media"

by Andrea Gallo, Leonardo Magherini, Federico Mondino, Carlo Bianco*, Tiziana Tosco, Rajandrea Sethi

In this document we present our point-by-point reply to the reviewers' comments. The comments are reported in in regular typeface (blue), whereas our responses are written in regular typeface (black). Line numbers refer to the revised version of the manuscript.

RESPONSE TO REVIEWERS

We sincerely thank the reviewers for carefully reading the work and for their constructive criticisms and valuable comments. Below, you will find a point-by-point description of each question and how each comment was addressed in the manuscript. The original reviewer's comments are reported in regular typeface (blue), our responses in regular typeface (black), and the modified manuscript text in red. Line numbers refer to the revised version of the manuscript.

REVIEWER #1

1. The Graphical Abstract should be revised to better reflect the current manuscript. In particular, the presentation of nZVI and Fe0 is not fully consistent with the revised text. Since the manuscript has replaced nZVI with nRIM, the Graphical Abstract should be updated accordingly.

   Response: We agree with the reviewer and thank them for pointing this out. The Graphical Abstract has been revised to better reflect the terminology and content of the current manuscript. In particular, the previous references to nZVI and Fe^0 have been replaced or revised consistently with the updated notation used in the manuscript. As also suggested by the reviewer in another comment, we have checked the entire manuscript and harmonized the terminology throughout the text, figures, and graphical materials.

2. Please standardize the terminology throughout the manuscript. At present, both "nZVI-based particles" and "nanoscopic Reactive Iron Minerals" are used. The authors should clearly define which term is preferred and use it consistently throughout the text, figures, and supplementary materials. In my view, "nZVI-based particles" may not be the most appropriate term, because the current evidence does not allow the authors to determine how much nZVI is actually present in the material.

   Response: We thank the reviewer for this valuable comment. We agree that the terminology should be standardized throughout the manuscript and that the term "nZVI-based particles" may be misleading. Although the presence of nZVI in the generated material is supported by the available evidence, we do not have sufficient information to quantify its actual fraction within the iron-based particles. To avoid

overinterpretation and ensure consistency, we have revised the terminology throughout the manuscript and now use the term “iron-based reactive nanoparticles” consistently.

3. The manuscript would benefit from clarification of the degradation products of PCE and TCE by nRIM. This information may help elucidate the dechlorination pathway and provide useful clues regarding the reactive components and degradation mechanism of nRIM.

Response: We thank the reviewer for this insightful comment. We agree that information on the degradation products of PCE and TCE would be valuable to elucidate the dechlorination pathway and to better identify the reactive components and degradation mechanism of the iron-based reactive nanoparticles. Unfortunately, degradation by-products were not analyzed during the experiments; therefore, the available dataset does not allow us to draw conclusions on the specific degradation pathways involved. However, we recognize that the identification and quantification of degradation products could be a relevant topic for future studies.

REVIEWER #2

1. Authors were able to address all comments to improve the manuscript, thank you. No further suggestions.

   Response: We thank the reviewer for their positive assessment of the revised manuscript and for acknowledging that the previous comments have been adequately addressed. We appreciate the reviewer's constructive feedback, which helped improve the quality and clarity of the manuscript.

Reviewer comments:

The manuscript entitled *In-situ synthesis of iron-based reactive nanoparticles for groundwater remediation: controlled emplacement in 3D and heterogeneous 2D porous media* by Gallo and colleagues presents a novel strategy for the in-situ synthesis of nZVI particles directly within a contaminated aquifer. Different protocols were employed for the synthesis of nZVI particles. An experiment was conducted in a 3D medium-scale homogeneous permeable aquifer. The authors used a ferrous chloride solution (named solution S) and an in-situ iron liquid (named solution R) to promote the nZVI synthesis. The authors also proposed 2D laboratory experiments replicating different heterogeneous aquifers with different arrangements of low-permeability layers. nZVI technologies are of interest for the remediation industry to effectively remove various groundwater contaminants. In particular, this manuscript focused on chloroethenes, which are a family of groundwater contaminants which remain of concern due to their complex (multiphase) transport and reactions in aquifers. The article proposes a novel approach for advancing knowledge on in-situ remediation technologies. While the authors demonstrated that nZVI were synthetized under different conditions in the aquifers, there are various aspects the authors should clarify to meet the criteria for publishing in Communications Earth & Environment.

Major comments:

1. The abstract is misleading in lines 34-35. Authors performed batch experiments with nZVI synthesized in the 3D aquifer. This sentence makes the reader believe that chloroethenes (PCE and TCE) were removed within 21 days in-situ, suggesting the technology could effectively be used for nanoremediation. Authors tested that the nZVI produced in the 3D aquifer can remove these compounds in batch experiments only (discussed in sections 2.3.4 and SM6). Authors did not attempt to inject chloroethenes into the 3D or 2D aquifers, therefore the reader is left without an explanation on why the reactivity of the nZVI produced in the aquifer was not tested under flow conditions. Proving that nZVI can be synthesized and effectively reduce a groundwater plume (whether in the 3D or 2D setup) would have been an important contribution. Authors should clarify why treating a contaminant plume in-situ was therefore not tested. This was only mentioned at the end of the manuscript (after lines 512) as a perspective. The 3D model aquifer has various piezometers that could have been used to track the contaminant plume and validate the approach. Geochemical interpretation as in Figure SM8 would have been possible.

2. The batch experiment showed that the removal efficiency of the produced nZVI in the 3D aquifer was between 30 and 40% for PCE and TCE, respectively (lines 441-443 and SM7). This removal efficiency is low. Why did authors choose a parallel test with nZVI produced with a batch synthesis protocol at 1 g/L and not at 0.1 g/L as formed in the 3D aquifers (Figure SM7)? What is the reasoning of testing concentrations at 20 and 10 ppm of TCE and PCE, respectively?

3. Authors validated the synthesis of the nZVI in a 3D medium-scale homogeneous aquifer using a tomography-like approach, which showed the production of nZVI around the injection well. A mixture of ferrous chloride, tap or deionized water and an iron liquid solution was induced.

Authors mentioned (lines 127-133) that the in-situ iron liquid consists of a blend of various reducing agents, including sulfates, sulfites, bisulfites, metabisulfites, dithionites, dithionates, thiosulfates, and pH modifiers. However, the reader is left without a proper understanding of how this iron liquid solution works, what the ratios and quantities in this solution are (how much is injected?), and so how this can be reproducible. One wonders: why not provide the exact composition of the solution? Authors provided the chemical characterization of the tap water (SM1), the injected concentration of the ferrous chloride but not much information on this iron liquid solution which is key for the production of nZVI. Is the solution commercially available? If not, it is not indicated in the manuscript if there is a confidentiality about the iron liquid solution. Could authors be more explicit about the composition of this solution and the concentrations injected? This would be of interest to the remediation industry who could test this approach and make the proper calculations (stoichiometry) for the synthesis of nZVI and projected contaminant removal.

Later in the text (Table 1) we read about the duration of the injection steps and cycles for solutions R, TW (water) and S in the 3D aquifer, but without the concentration of solution R (the iron liquid). We read again something similar in section 2.4.3 for the 2D aquifers. In line 340, authors mentioned that their protocol resulted in the production of 160 mg of iron (assuming a yield of 100%) in the 2D aquifer. This iron liquid solution, composition, concentrations and the exact protocol of its injection should be clarified. This is necessary for scale up and for testing in other aquifer setups, pilot tests and real scenarios.

Minor comments:

In the abstract: Please consider removing environmentally relevant contaminants and mention chloroethenes.

Line 75: Please clarify the tendency of contaminants to accumulate in low-permeability layers. Contaminants are affected by different processes. Is it because of sorption, diffusion, density (DNAPL) or due to low-hydraulic conductivity? Is this particularly important for chloroethenes?

Line 99: Please clarify which natural reducing agents. This would help linking geochemical conditions in natural aquifers.

Line 175. The hydrodynamic parameters of the porous medium were obtained from column tracer tests performed **in** the same material .... I think it should be read **with** the same material. Tracer tests were not tested in the 3D or 2D setup. Authors should provide information on why tracer tests were not performed in the aquifers.

Line 238: When was the excavation process performed for the 3D aquifer and the tomography-like approach? Please clarify how many pore volumes passed so the reader can have an idea how long it took to produce the nZVI. Was the aquifer drained prior to the excavation?

Figure SM3: I recommend adding the distances x, y as similarly presented in Figure 3B.

Line 396: The deposition yield wouldn’t be between 60 and 94% for external and inner areas of the reactive zone, respectively ? Please check this sentence.