Assessing Biomineral Formation by Iron-oxidizing Bacteria (FeOB) in a Circumneutral Creek

NOHEMI ALMARAZ[1,2*], ANDREW H. WHITAKER[2], MEGAN Y. ANDREWS[2], and OWEN W. DUCKWORTH[2]
[1]Department of Civil & Environmental Engineering, Colorado School of Mines 1500 Illinois Street, Golden, CO 80401 USA (*presenting author)
[2]Department of Soil Science, North Carolina State University, PO Box 7619, Raleigh, North Carolina
*Email: nalmaraz@mines.edu

ABSTRACT

The role of microorganisms in the oxidation of iron is of critical importance to many scientific fields, and has real-world applications in soil remediation and water treatment. Iron oxidation is a common phenomenon associated with the discharge of anoxic, iron-rich groundwater into rivers and creeks where oxygen is readily available. Yet, neutrophilic iron-oxidizing bacteria (FeOB) exist primarily in locations with favorable redoxcline, circumneutral pH and high Fe (II) concentrations. Thus, environmental and water chemistry factors are major contributors to the composition and morphologies of each site.

INTRODUCTION

- Biomineral formation can be attributed to both biogenic and abiotic factors.
- FeOB live in the suboxic region with high Fe (II) concentrations.
- Ferric iron is insoluble at neutral pH.
- Iron biomineral has unique characteristics in the sorption of metals that make it a possible solution for emerging environmental problems.

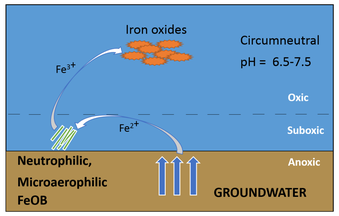


Figure 1. The biological process by which FeOB oxidize ferrous iron (Fe^{2+}) into ferric iron (Fe^{3+}) forming iron oxides.

OBJECTIVE

This study will elucidate the characteristics of various FeOB niches that contribute to the formation of biomineral. This will help improve the understanding on how water chemistry and bacterial communities affect biomineral formation. Thus, provide more information on the viability of biominerals for the removal of hazardous chemicals from water.

SITE OVERVIEW

Walnut Creek (WC)

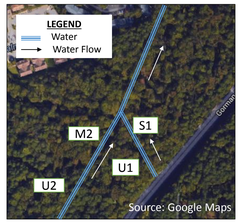
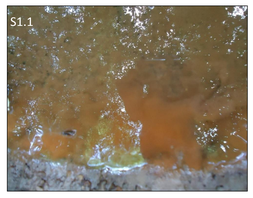


Figure 2. Sampling site characterized by the discharge of two streams into one stream. Different iron-oxidation abundance was observed through the three streams.

Site 1: Underground seep (S1) with biomineral formation.
Deep, slow moving water (U1) with no iron oxides.
Site 2: Running water with no visible iron oxides (M2).
Deep, slow moving water with no iron oxides (U2).



Pullen RBC (Continuous water current with iron-oxides along its banks)

Figure 3. Site E located downstream and in opposite side of the other 4 sites.

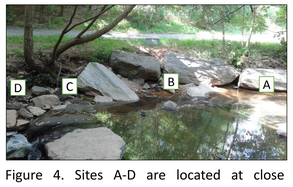
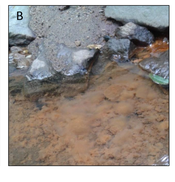
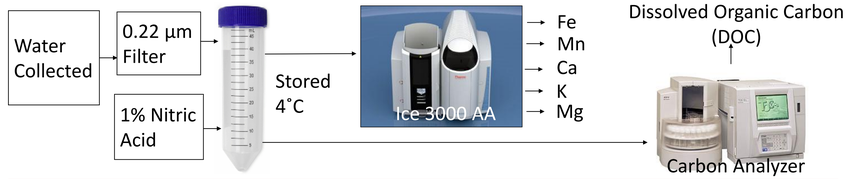


Figure 4. Sites A-D are located at close proximity of each other upstream of site E .

Site A & B : similar characteristics of biomass, lighter in color and similar appearance.
Site C & D: similar appearance, slimy texture and formation of sheen.
Site E: distinct appearance from all other sites, visually it had more dense formation of biomineral.



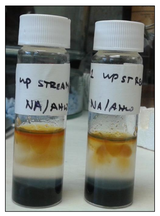
ANALYSIS OF WATER CHEMISTRY



BACTERIAL ISOLATION

Gradient Tube:
- Headspace
  - N_2 and CO_2
- Media layer
- Iron sulfide mineral plug
- Inoculated with 100 µL of biomass

Incubation:
- Room Temp.
- 4 weeks



Inoculation:
- Transfer 100 µL of sample to new tube
- Re-purge tube with N_2 and CO_2 and continue incubation
- Prepared microscope slides for imaging

EVOS XL Microscope

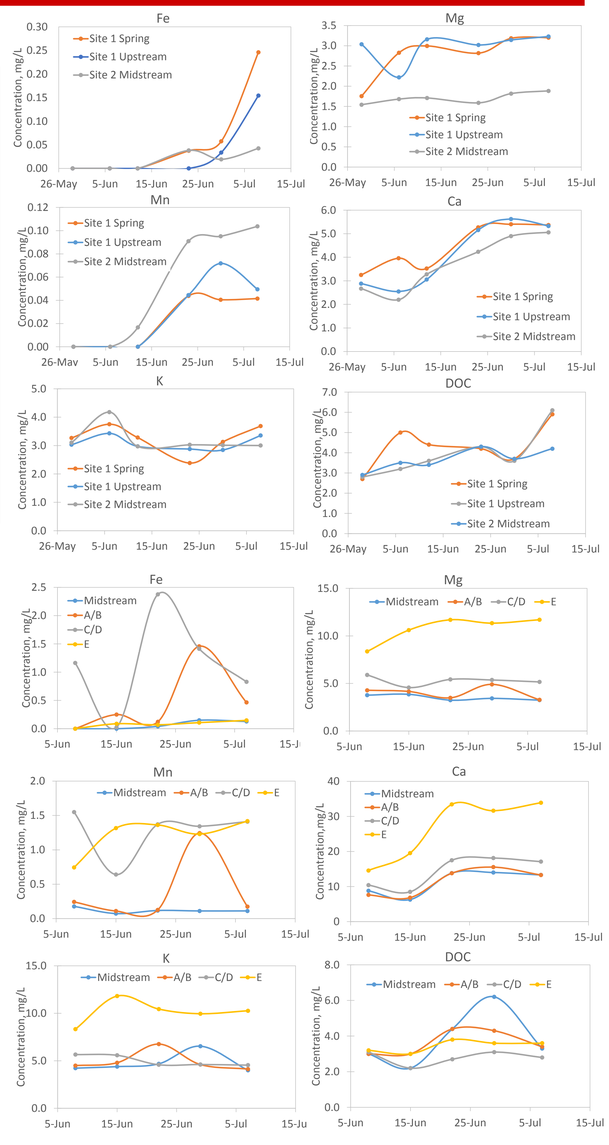
RESULTS

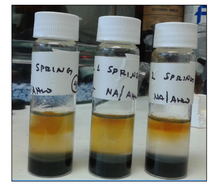
Walnut Creek

- Average Temperature: 24°C
- Average pH: 6.7
- **Fe** and **Mn** are present at lower levels than the other metals
- **Fe** and **Mn** increase throughout the summer
- All sites have similar trends in **Fe, Mn, Mg, Ca** and **K** levels possibly as a result of similar water source
- **Ca** and **Mn** have a similar trend

Pullen RBC

- Average Temperature: 24°C
- Average pH: 6.5
- **Fe** and **Mn** are present at lower levels than the other metals
- **Fe** and **Mn** have similar trend indicating geochemical mixing
- **Fe** and **Mn** in A/B and C/D have significant fluctuations
- The distinct difference in composition of E to the other sites indicates a different water source contributing to the formation of biominerals





- Red-brown (biological oxidation) band near the top
- Oxidation on top of the media closer to where oxygen is present
- Morphology: dark tubular structures

Figure 4. Bacteria from S1 three weeks after inoculation.

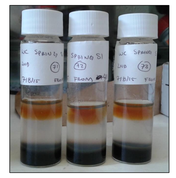
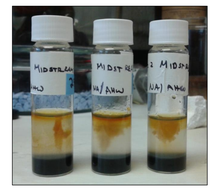


Figure 7. 2nd S1 inoculation 2 weeks after incubation.



- Site had no visible iron oxides
- Vertical formation of iron oxides (not banded)
- Irregular-shape of iron oxides
- Morphology: dense and round shaped solids

Figure 5. Bacteria from M1 three weeks after inoculation.

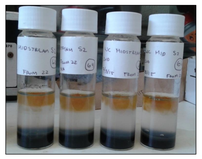
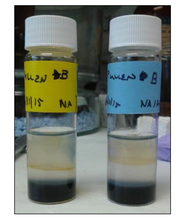


Figure 8. 2nd M1 inoculation 2 weeks after incubation.



- Light colored band
- Thin oxidation band in the middle of media
- Morphology: tubular structures as well as round "blobs"
- All tubes from Pullen RBC had a similar appearance

Figure 6. Bacteria from Pullen B three weeks after inoculation.

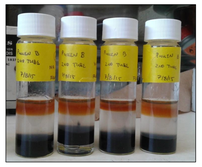


Figure 9. 2nd B inoculation 2 weeks after incubation.

DISCUSSION

- Despite the significant difference in concentration of most metals in the two sites, both had iron oxides growing on them
- There are distinguishable differences between sites A/B and C/D despite their close proximity
- In WC, levels of Mn and Fe were higher in deep, slow moving water
- The presence of iron oxides was variable throughout the summer but no specific trend for this occurrence was determined
- Formation of iron oxides on the top layer of the media may be an indication of FeOB that are less microaerophilic like *Leptothrix*
- Samples with no visible iron oxides on site grew iron oxides in the lab. This is possibly due to the presence of FeOB but not a sustainable environment for the formation of biominerals
- Distinct morphologies are observed through different sites in the gradient tubes as well as the images
- The second inoculation of bacteria from an existing tube resulted in faster and different formation of iron oxides

CONCLUDING REMARKS

- Water of varied chemistries can support FeOB and biomineral formation
- Biomineral can form in a variety of ways, the exact factors limiting its formation are yet to be determined
- Ongoing work will phylogenically determine the FeOB present at each site

ACKNOWLEDGMENTS

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