

Background

The interaction between crustal rocks rich in iron (e.g., basalts, peridotites) and water generates hydrogen (H₂) via several processes collectively known as serpentinization. These reactions are widespread and were likely active early during Earth's history; comparable processes can be expected on other rocky planets that have liquid water. The H₂ produced through these water-rock interactions likely had an integral role in supporting ancient chemoautotrophic metabolisms.

The influence of lithogenic H₂ on the distribution of H₂-dependent chemoautotrophic metabolisms and the microbial communities they support can be explored through the integration of geochemical, molecular, and physiological data in modern early earth analog systems. In particular, such relationships can be explored in the accessible hot springs in Yellowstone National Park (YNP) (Fig. 1) which provide a range of lithogenic H₂ sources that have been suggested to support diverse H₂ metabolizing microbial communities. Gradients in oxidant availability due to differences in subsurface geology (Fig. 2) may dictate the nature of H₂ metabolisms in geothermal communities in YNP.

We hypothesize that chemotrophic organisms involved in H₂ metabolism have diversified to capitalize on available gradients in oxidants

- Goal 1:** Integrate geochemical, molecular and physiological approaches to link populations with chemotrophic H₂ transformation activities in YNP hot springs
- Goal 2:** Determine the extent to which H₂ metabolic activity is coupled with various oxidants and their availability (e.g. CO₂, SO₄, NO₃, O₂)
- Goal 3:** Quantify the extent to which distribution and diversity of hydrogenase genes reflects availability of oxidants

Site Location

Sampling Locations: **Roadside West (RW) Spring** and **Roadside East (RE)** (Fig. 3, Fig. 4) are a paired set of hot springs (Fig. 2) which were chosen because they are likely to exhibit differences in the availability of H₂-metabolism supporting metabolisms.

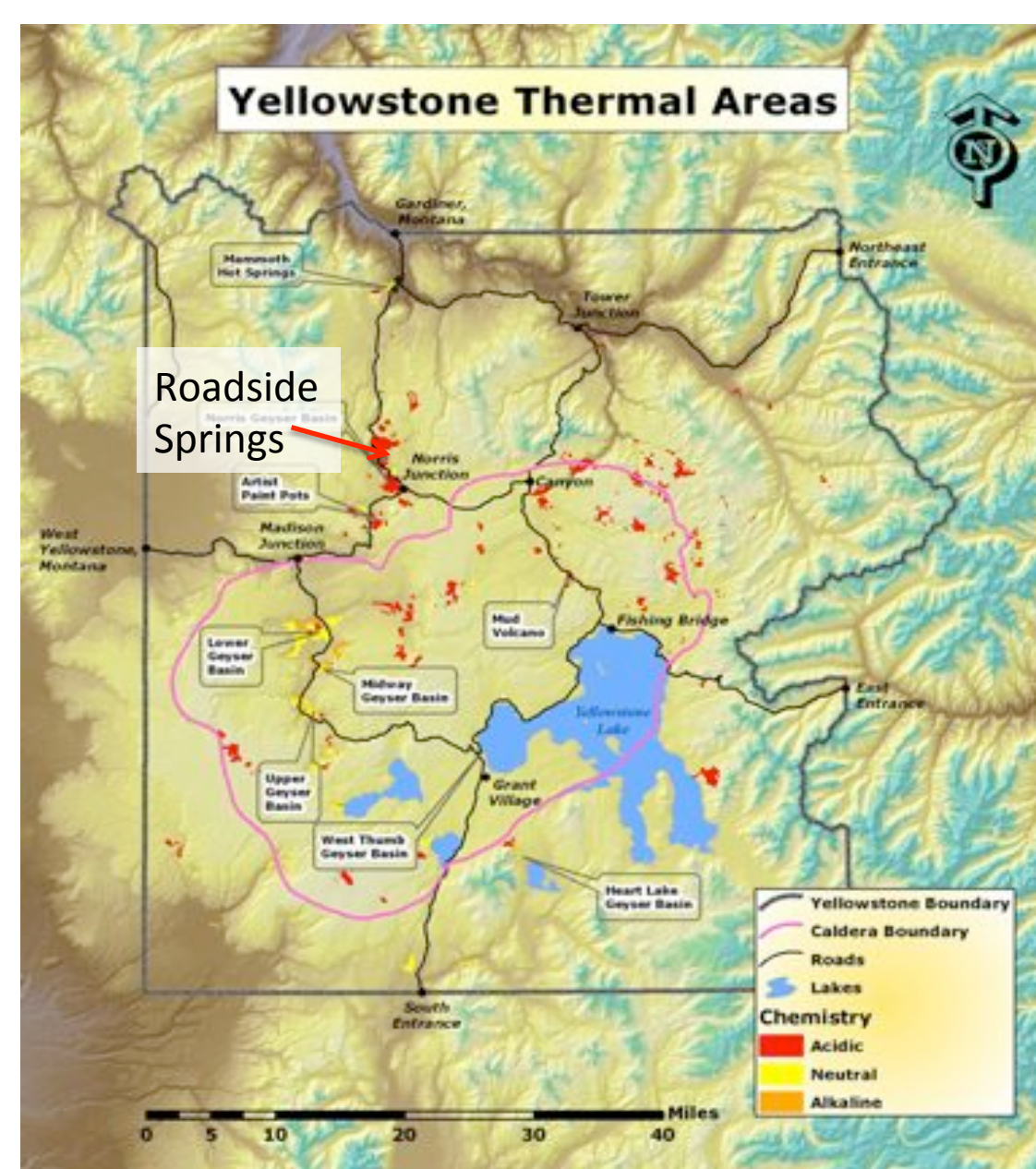


Figure 1. Location of Roadside Springs in YNP.

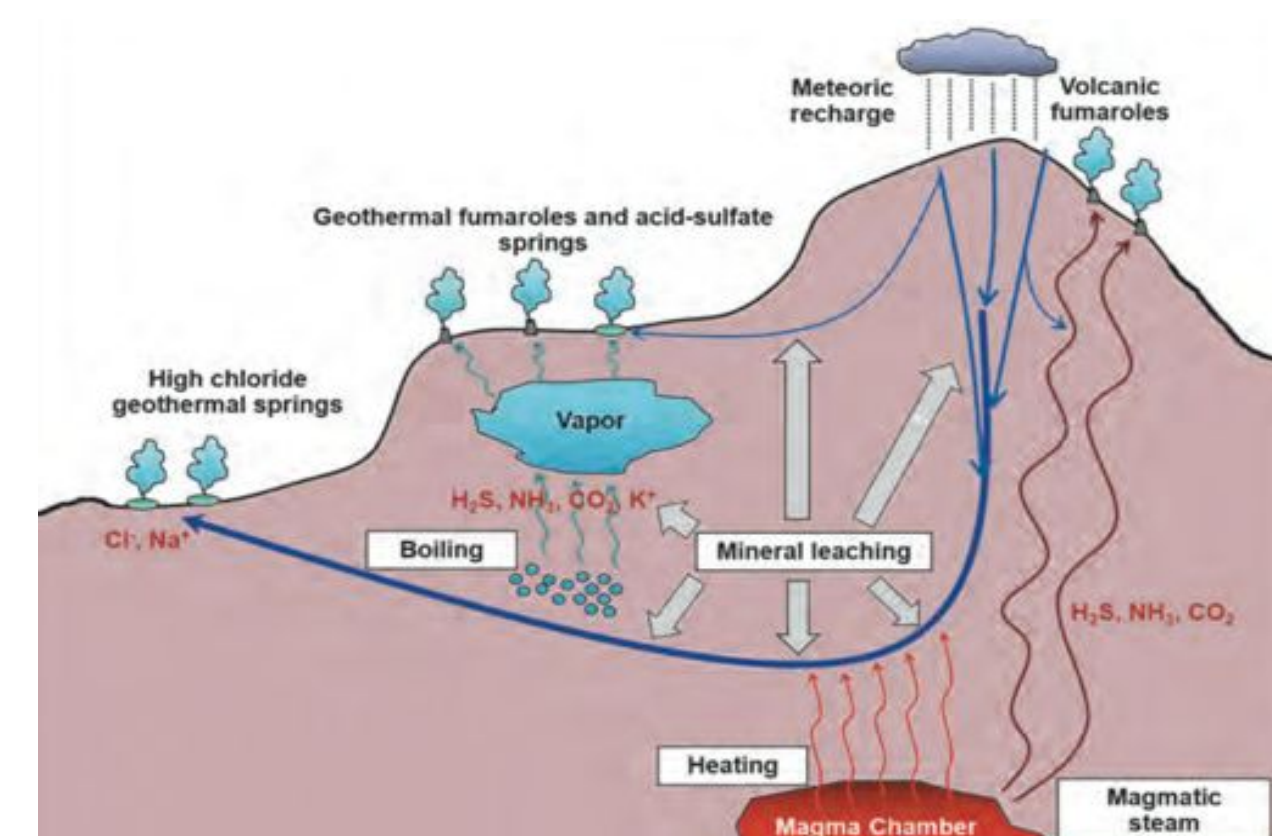


Figure 2. Model of subsurface phase separation leading to acidic and alkaline springs.

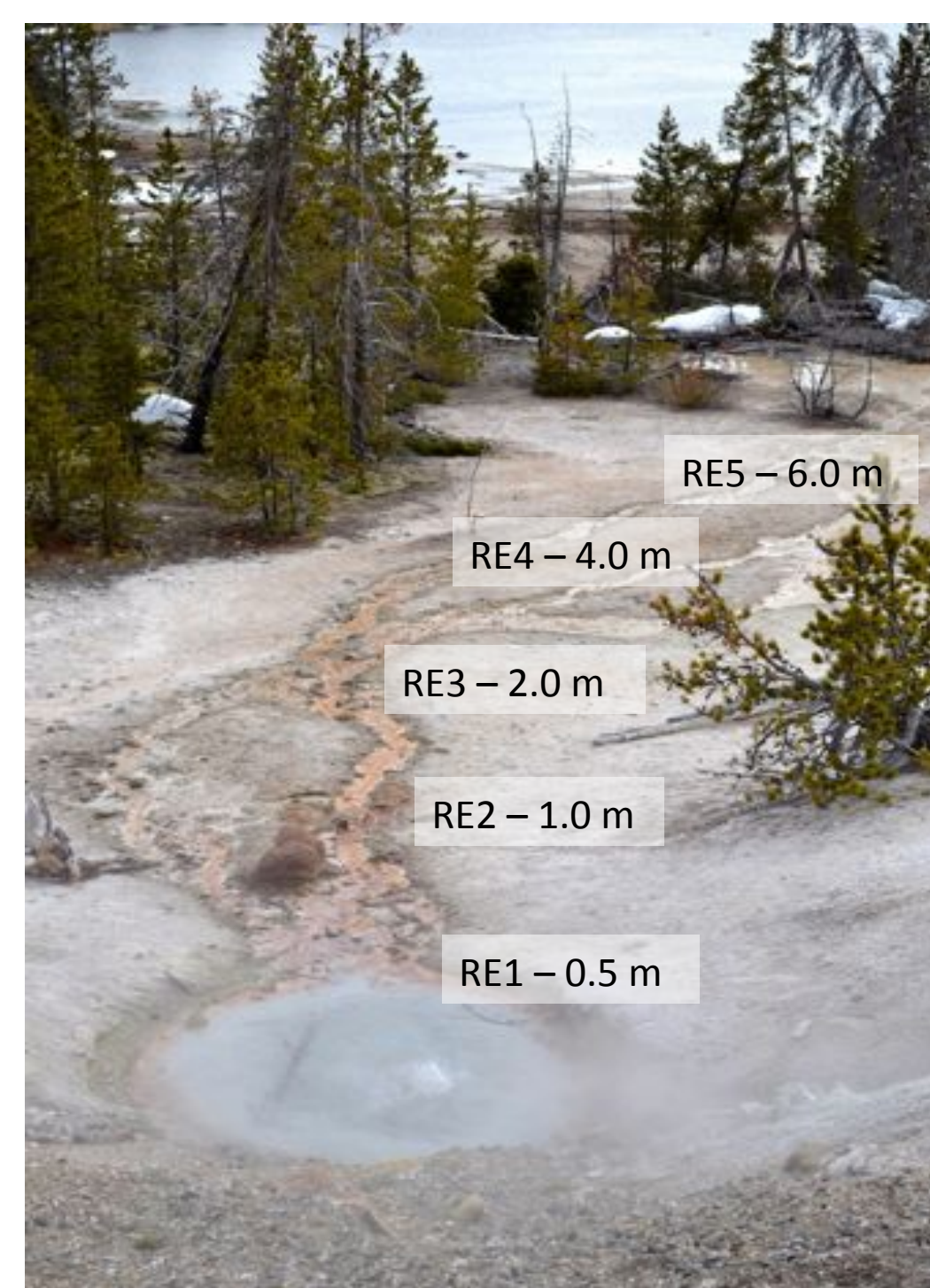


Figure 4. RE spring source and outflow channel

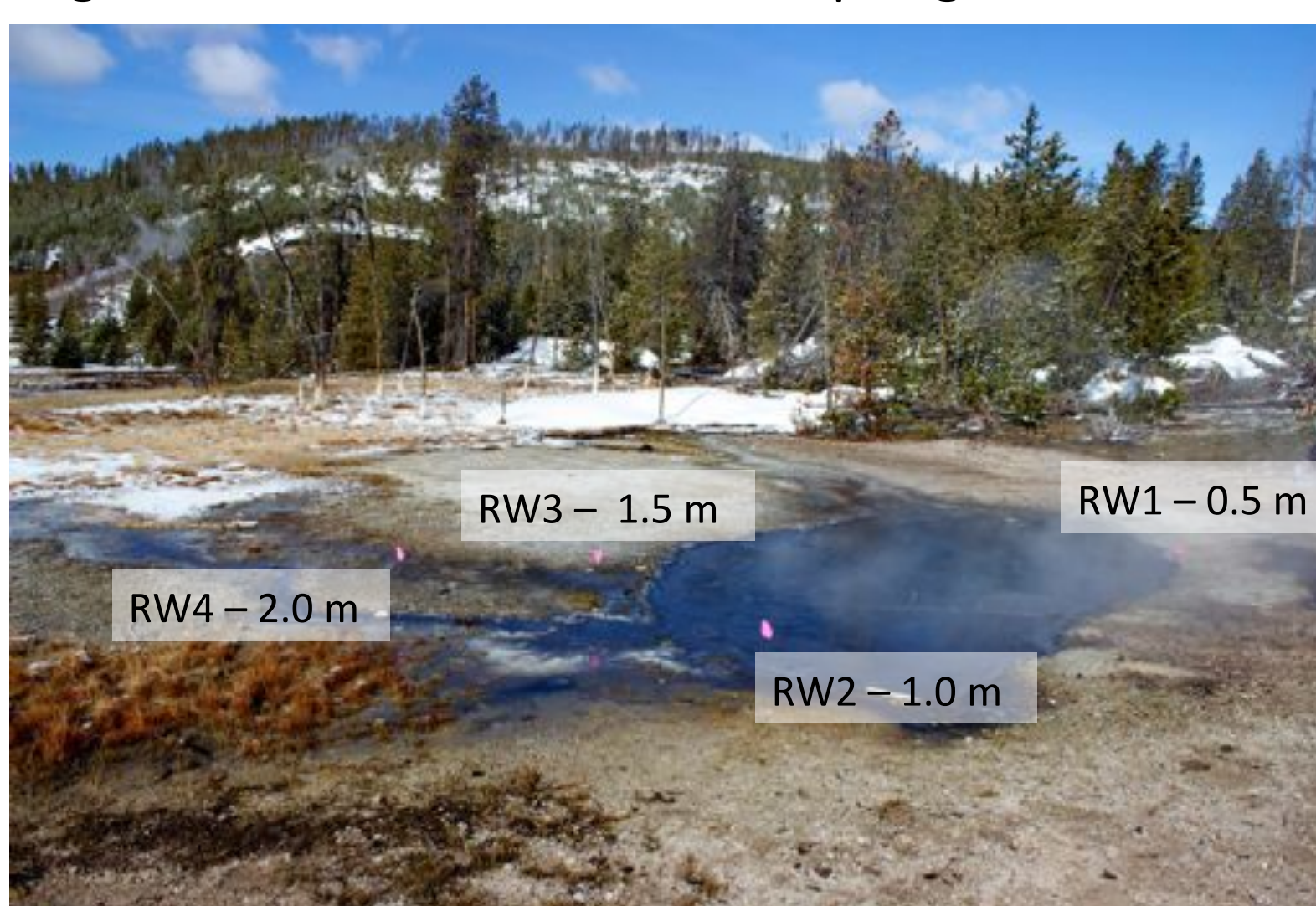


Figure 3. RW spring source and outflow channel

Methods

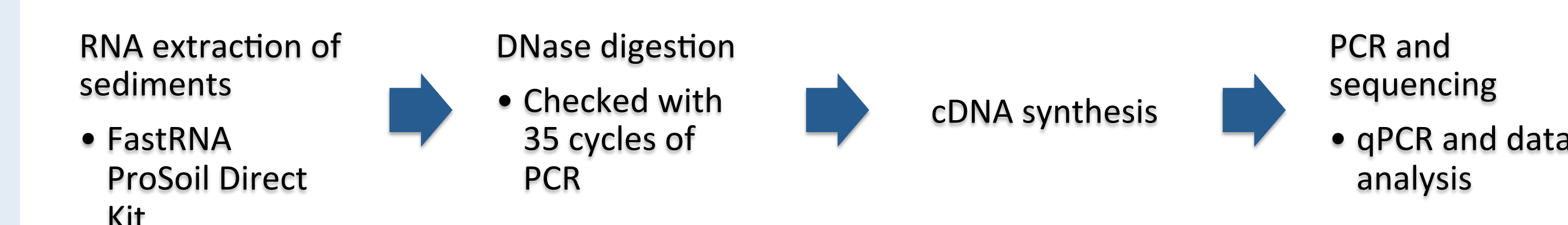


Figure 5. Molecular analysis workflow

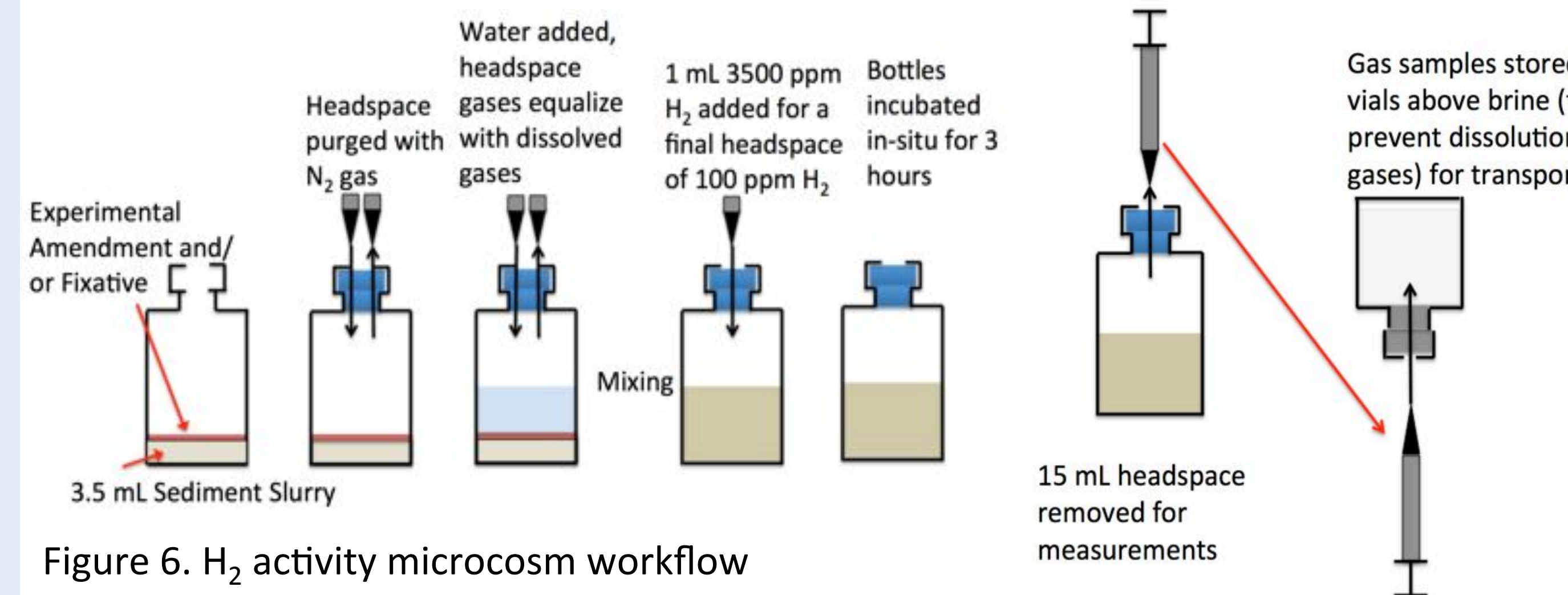


Figure 6. H₂ activity microcosm workflow

Results – Aqueous and Gas Phase Geochemistry

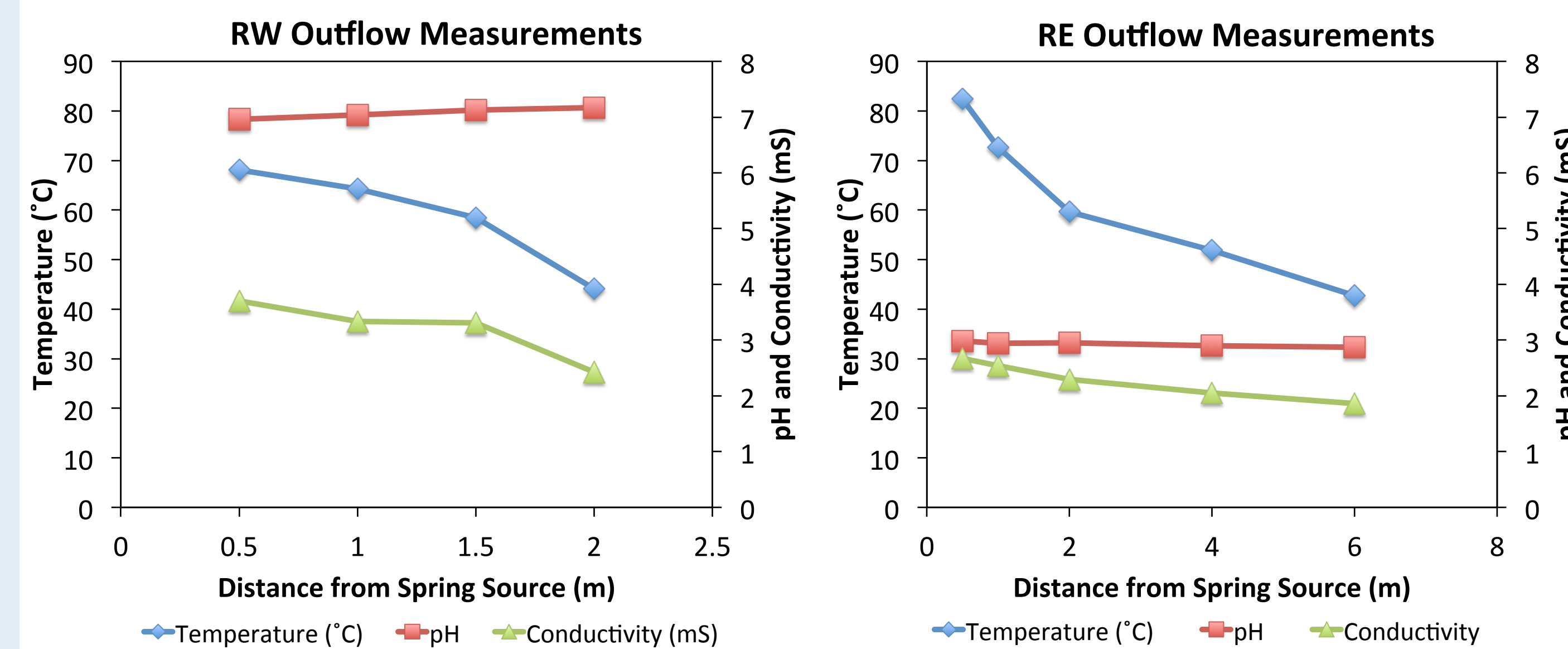


Figure 7. Fluid temperature, pH and conductivity measured down RW and RE outflow channels.

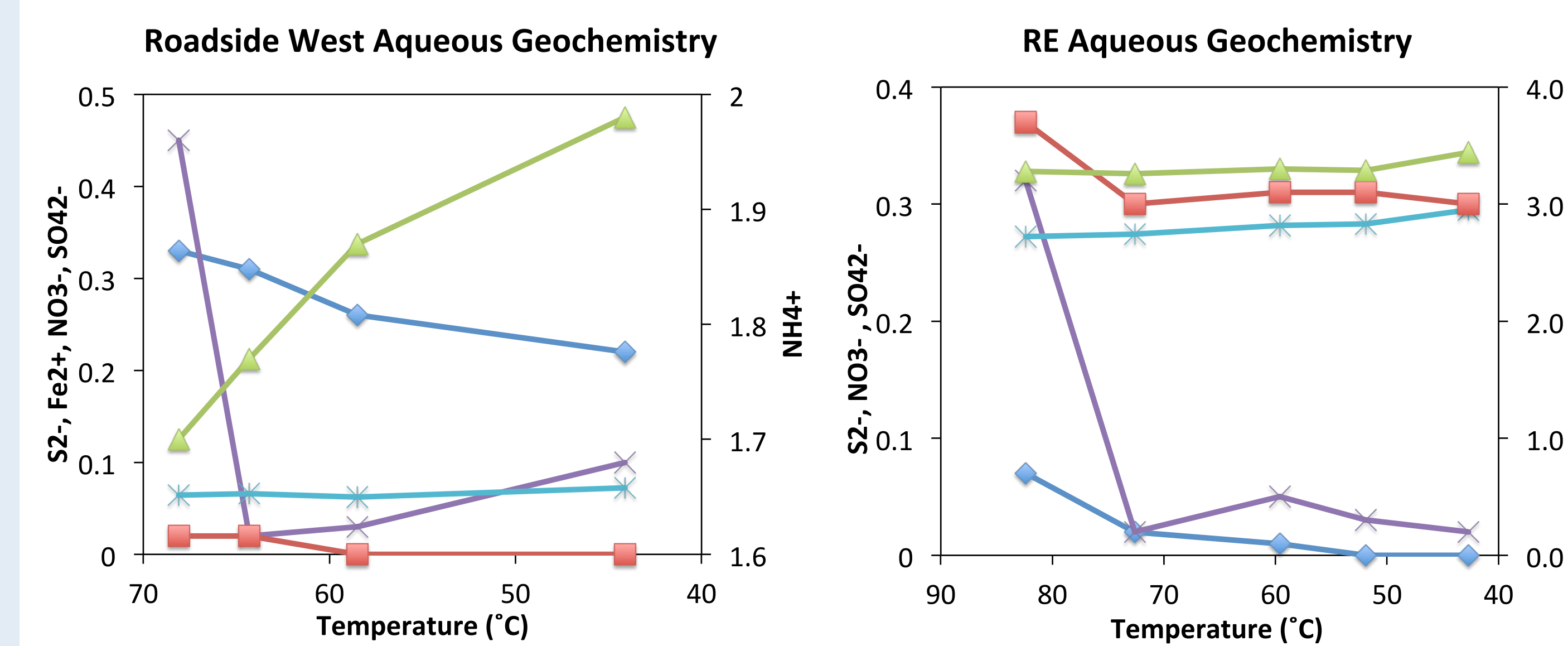


Figure 8. Geochemical gradients; specifically sulfide, iron, nitrate, sulfate, and ammonia as measured down RW and RE outflow channels.

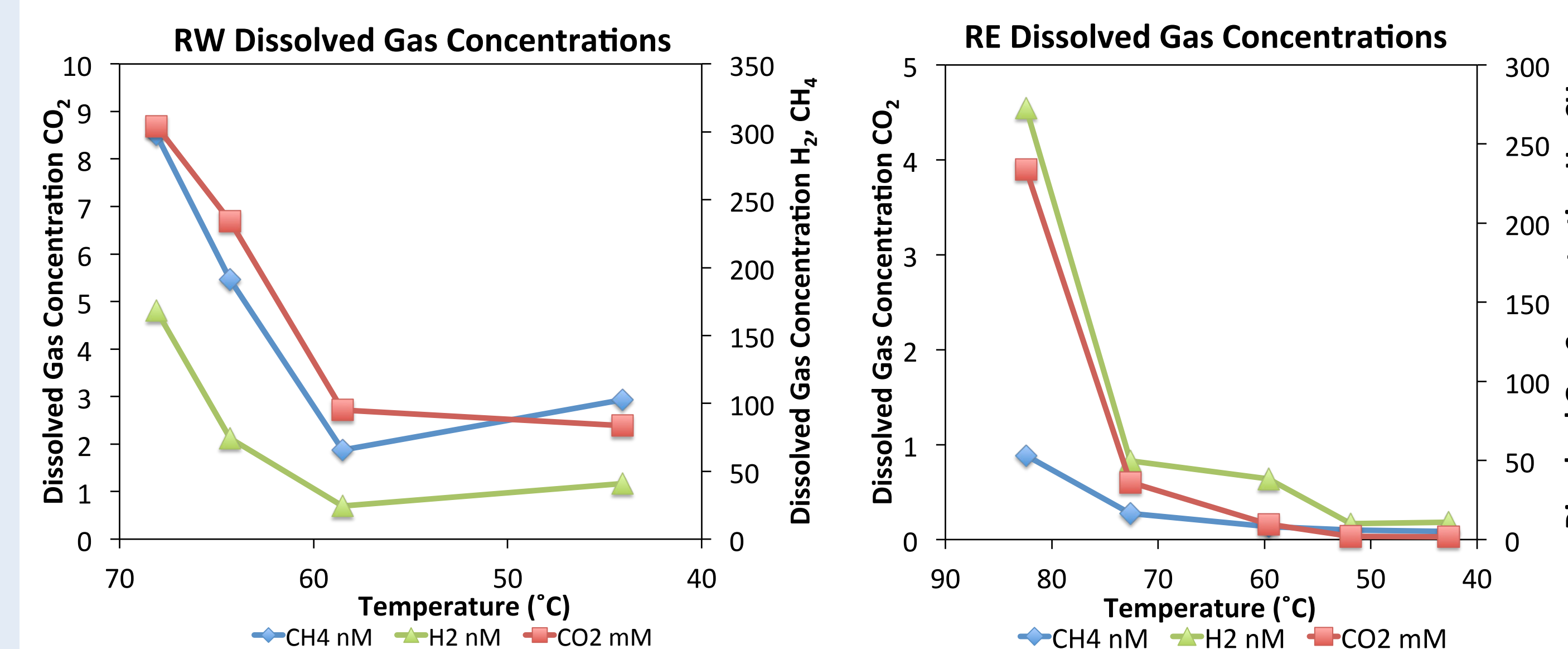


Figure 9. Gradients in dissolved gases (CH₄, CO₂, H₂) along the RW and RE outflow channels.

Results – Microbial Activities

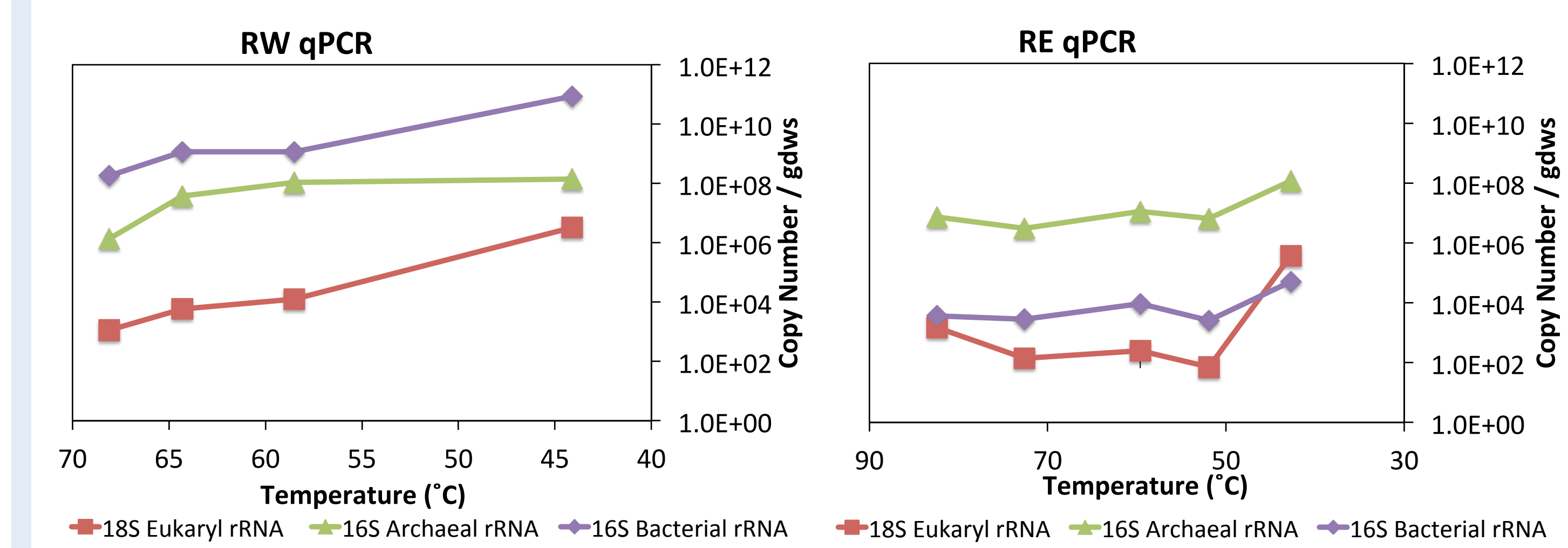


Figure 10. Abundance of bacterial, archaeal, and eukaryal SSU rRNA transcripts in the RW and RE outflow channels.

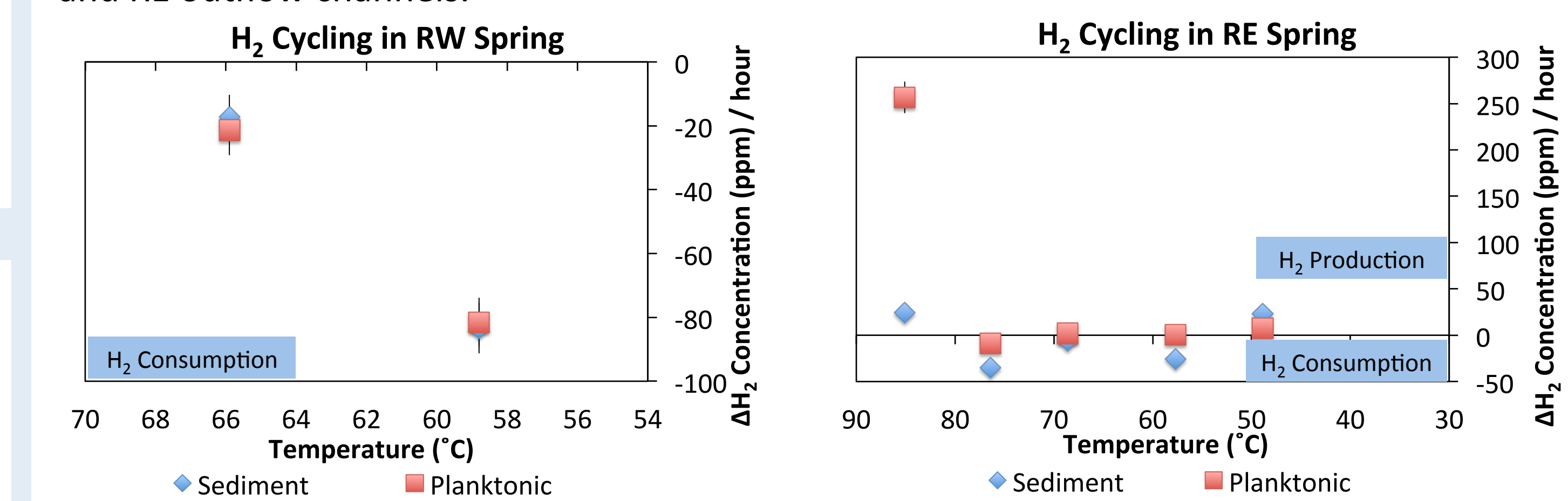


Figure 11. Net change in H₂ concentration during a 1 hour incubation, indicating H₂ production or H₂ consumption in planktonic and sediment-associated microbial communities.

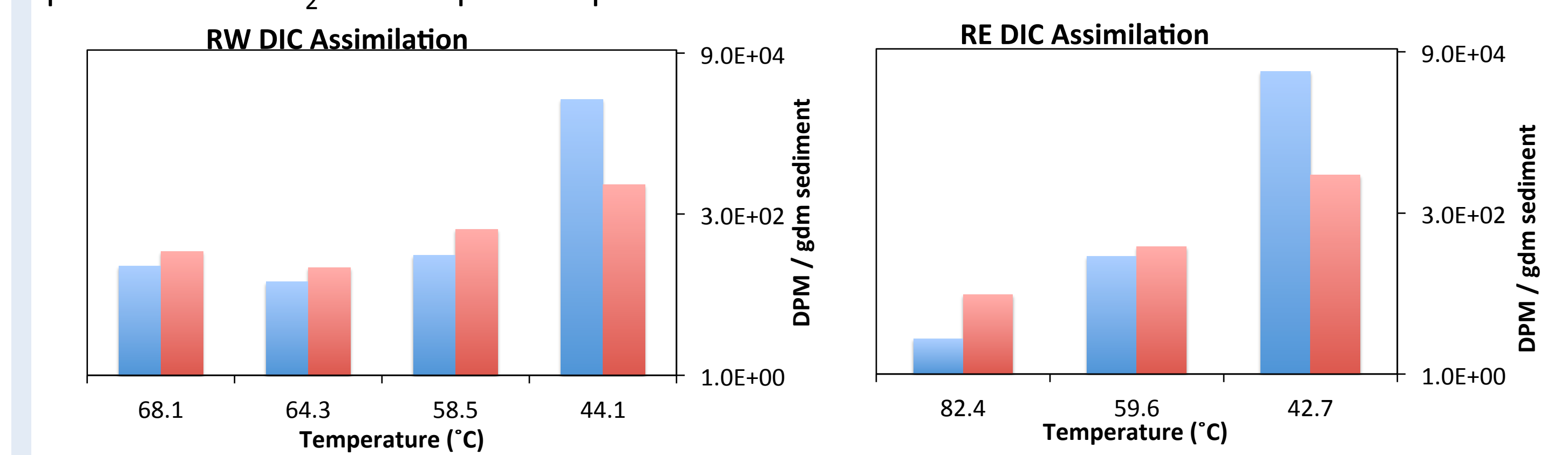


Figure 12. Dissolved Inorganic Carbon (DIC) assimilation in sediment-associated communities from RW and RE incubated at different temperatures along the outflow channel.

Conclusions

- SSU rRNA transcript abundance increases as temperature decreases and as pH becomes more neutral, which may reflect decreased maintenance energy demands at lower temperature or more circumneutral pH
- Decreasing levels of some substrates (e.g. NO₃⁻) down RE and RW outflow channels (Fig. 8) could reveal the preferred e⁻ acceptors coupled with H₂ oxidation.
- While most communities were net sinks for H₂, one (RE1) was a net source of H₂ suggesting a role for biology in producing H₂ in hot spring communities
- The abundance of the bacterial 16S rRNA transcripts along the RW outflow channel correlates with net H₂ consumption activities, while the abundance of archaeal 16S rRNA transcripts along the RE outflow channel suggests that archaea have a role in net H₂ production activities.
- Rates of CO₂ fixation and the abundance of SSU transcripts are correlated and exhibit a marked increase as the communities transition from chemotrophic to phototrophic metabolisms.

Future/Ongoing Work

- Ongoing multiple probable number assays and sequencing of SSU rRNA and hydrogenase transcripts will provide insight into the preferred redox partner(s) for H₂ metabolisms, the identity of the active species in these hot spring microbial communities, and the activity and identity of hydrogenases responsible for H₂ metabolism in YNP hot springs.
- Phylogenetic analyses of SSU rRNA and hydrogenase transcripts when integrated with geochemical and physiological assays will provide a mechanism to examine the extent to which the diversification of hydrogenases and H₂-cycling microbial communities reflect available energy gradients in this early earth analog system.