Tanbark Creek Year 2 (2024-2025) Long-Term PRB Performance Monitoring

1/29/26

Submitted to:

Peconic Estuary Partnership
Director Joyce Novak, PhD

Submitted by:

Molly Graffam, PhD
Cornell Cooperative Extension of Suffolk County

with data provided by
CLEAR
Ron Paulsen, PG
Patrick Murray

Abstract:

Cornell Cooperative Extension of Suffolk County (CCE) completed the 2nd year of quarterly monitoring (2024-2025) of a pilot injection-style permeable reactive barrier (PRB) at Tanbark Creek in Three Mile Harbor. In year 2, nitrate upgradient of the pilot PRB (untreated groundwater) was on average 1 mg N/L higher than was measured in year 1. Untreated groundwater nitrate was moderately elevated during the monitoring period and ranged from <0.3 to 7.51 mg N/L. According to Wilcoxon rank-sum tests, nitrate concentrations were significantly lower in downgradient wells (treated groundwater) compared to upgradient wells (untreated groundwater) across all seasons. On average we measured 72% nitrate removal corresponding to 48-126 lbs N of nitrate removed before it could enter Tanbark Creek. Despite higher average nitrate in untreated groundwater, the original 5 porewater stations had the lowest average nitrate measured thus far in spring 2025 (data provided by CLEAR). Year 2 results were communicated with Peconic Estuary Partnership Management and Policy Committee and the Town of East Hampton Water Quality Technical Advisory Committee.

Introduction:

The Tanbark Creek PRB is a pilot project intended to demonstrate the use of carbon emulsion injection for groundwater nitrate reduction. The installation of injection wells and the injection of the carbon emulsion concluded at the end of 2022. Post-installation research to determine the effectiveness of the current design and learn ways to improve upon it began in summer 2023. This project intends to add to the repository of PRB performance data and inform several long-term goals for PRB implementation and specifically the carbon injection installation approach. Long term project goals include relating inland PRB performance to offshore porewater and surface water quality, varying the carbon amendment solution strength to minimize the installation time, quantifying the PRB longevity, characterizing the redox reactions downgradient of the PRB, and maximizing nitrate reduction by potentially including other environmentally approved amendments along with EOS-100 to support the proper conditions and/or microbial community.

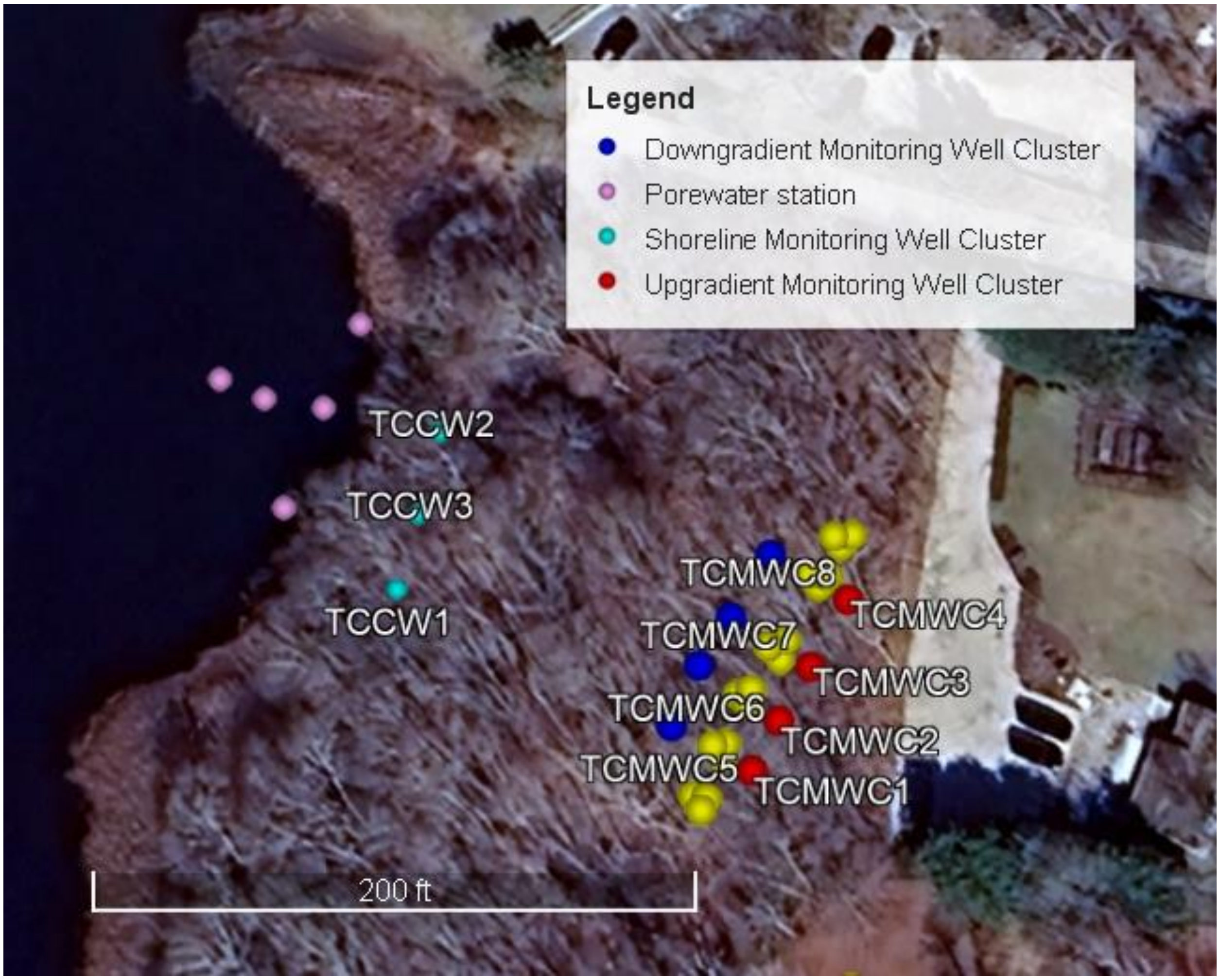


Figure 1: Tanbark Creek site schematic showing location of pilot PRB injection wells, upgradient and downgradient monitoring well clusters (TCMWC1-8). There are six injection clusters starting with TCINJ1 in the south and moving northeast to TCINJ6. The red and blue dots indicate 2 wells within 1 ft of each other screened at 20-25 ft and 10-15 ft below grade. Light blue dots represent shoreline cluster comprised of 3 wells screened at 5-15 ft, 15-25 ft, and 25-35 ft below grade. Light purple dots represent the 5 original long term monitoring offshore porewater stations.

Methods:

A. PRB performance monitoring

Four upgradient PRB performance monitoring well clusters (TCMWC1-4; untreated groundwater) and 4 downgradient well clusters (TCMWC5-8; treated water) (2 depth intervals per cluster; 16 wells total) were sampled for nitrate, nitrite, ammonium, and TKN in fall 2024, winter 2025, spring 2025, and summer 2025 (Fig. 1). Fall 2024, winter 2025, spring 2025, and summer 2025 samples were collected on 10/17/24, 2/3/25, 3/31/25, and between 6/25/25 and 6/27/25, respectively. Additionally, performance monitoring samples were analyzed for sulfate, hydrogen sulfide, alkalinity, dissolved organic carbon (DOC) and dissolved iron during the summer 2025 sampling event. Hydrogen sulfide is defined as the sum of H_2S and HS^- present in the sample after the laboratory stabilized the pH at 7. Shoreline cluster wells were sampled on 10/31/24 and 11/5/24 for nitrogen series, dissolved iron, and DOC.

PRB performance monitoring wells were purged of three well volumes prior to sampling using a peristaltic pump running at approximately 500 mL/min. Samples were collected for laboratory analysis when field parameters including conductivity, total dissolved solids (TDS), oxidation reduction potential (ORP), pH, and temperature were stable (±10% relative standard deviation (RSD)). Conductivity, TDS, ORP, pH, and temperature were measured onsite at the time of collection with a Myron Ultrameter. Dissolved oxygen (DO) was measured with a YSI DO meter. Depth to water was measured with a Solinst water level sounder.

Total Nitrogen (TN) concentration is the sum of TKN and nitrate. Nitrite was not included due to values always below the limit of quantitation (LOQ). For values below the LOQ, half the LOQ was used for calculating average and standard deviation for nitrate, ammonium, and TKN.

B. PRB carbon injection longevity monitoring

Soil borings were collected 2-10 ft away from the pilot PRB injection wells and 2 ft away from upgradient PRB monitoring wells at 20-25 ft below grade on 9/10/25. More specifically, samples were collected at similar locations last year near PRB injection well clusters 2, 4, and 5 (TCINJ2, TCINJ4, TCINJ5) and near upgradient monitoring well clusters (TCMWC1, TCMWC 3, and TCMWC4). Soybean oil analysis was done at Long Island Analytical laboratory using FID instrument and chromatography method. This test could detect presence or absence of soybean oil. Replicates within the PRB zone of influence and outside the expected zone of influence were collected to validate the anticipated presence/absence of soybean oil which is the primary carbon source in the PRB. Soil samples were also analyzed for total organic carbon (TOC) as another approach to validate and quantify the presence of organic carbon.

C. Tanbark Creek water quality monitoring

Porewater, surface water, and submarine groundwater discharge (SGD) rates were previously measured in 2020/2021 prior to PRB installation. Each year, CCE's subcontractor Coastline Evaluation Inc. (dba CLEAR) returns to the same pre-PRB installation stations. However, this year the offshore porewater work also included an expanded grid of stations. For the original 5 porewater stations, three porewater samples were collected along the shoreline and two additional samples were collected approximately 25 ft and 50 ft west of the shoreline to form an offshore transect (Fig. 1). Porewater sampling is accomplished using a patented porewater evaluation instrument called the Trident Probe provided by CLEAR. The probe uses a direct-push approach and is deployed by boat. The probe also simultaneously collected in-situ temperature and conductivity readings. Specifically, the Trident Probe measures temperature and bulk conductivity contrast between porewater 1 ft below the sediment-water interface and surface water 1 ft above the sediment-water interface. Areas with high temperature contrast and low porewater conductivity are conditions indicative of groundwater discharge. A seepage meter was deployed at 2 stations; close to the shoreline (TCSP7_5) and approximately 25 ft from the shoreline (TCSP7_25) to quantify SGD flow rate over a tidal cycle. The seepage meter is calibrated, and a zero check is done during the deployment at each station by collecting a "zero" reading with the outflow tube closed. Surface water grab samples were collected by hand from the middle of the water column just above the porewater stations and porewater samples were collected using a low-flow peristaltic pump. A minimum of 200 mL was purged prior to collection of a porewater sample.

Results and Discussion:

Quarterly nitrate data from year 1 is presented in Figure 2 for comparison with this year's quarterly data in Figure 3. Overall, in year 2 Wilcoxon rank-sum tests indicated that nitrate concentrations were significantly lower in downgradient wells (treated groundwater) compared to upgradient wells (untreated groundwater) across all seasons (details provided in Exhibit A).

A. Fall 2024 PRB Performance monitoring

Groundwater conductivity ranged from 246-460 µS/cm and average groundwater temperature was 14.1 °C during the fall sampling event. Groundwater nitrogen consisted of nitrate, ammonium, and organic nitrogen. Nitrite was always below the LOQ. Upgradient wells at 10-15 ft and 20-25 ft below grade had 3.0 ±0.9 mg N/L and 5.6 ±2.0 mg N/L as nitrate, respectively. Downgradient wells at 10-15 ft and 20-25 ft below grade had 0.3 ±0.1 mg N/L and 1.8 ±1.5 mg N/L as nitrate, respectively (Fig. 3).

In fall of 2024 shoreline cluster wells (Fig. 1; light blue dots) were sampled for dissolved iron and DOC, and below detectable levels were measured for both analytes in all shoreline wells screened from 5 to 35 ft below ground. The shoreline cluster wells are approximately 100-115 ft west (further downgradient) of the downgradient PRB monitoring wells. Our data is consistent with Borden and Lieberman 2009, indicating that approximately 100 ft or less is sufficient distance for complete redox recovery. In the future, a transect of wells at multiple distances away from the PRB can further elucidate the redox reactions occurring downgradient of the barrier and the exact distance to achieve complete reprecipitation.

B. Winter 2025 PRB Performance Monitoring

Groundwater conductivity ranged from 246-479 µS/cm and average groundwater temperature was 11.4 °C during the winter sampling event. Groundwater nitrogen consisted of nitrate, ammonium, and organic nitrogen. Nitrite was below the LOQ in all samples. Similar to fall, all upgradient monitoring wells at 20-25 ft below grade had higher nitrate than 10-15ft below grade. The largest difference was observed at TCMW4 where there was 6.56 mg N/L at 20-25 ft and 3.12 mg N/L at 10-15 ft. Upgradient wells at 10-15 ft and 20-25 ft below grade had 2.4 ±0.8 mg N/L and 4.9 ±1.8 mg N/L as nitrate, respectively. Downgradient wells at 10-15 ft and 20-25 ft below grade had 0.3 ±0.2 mg N/L and 1.9 ±1.7 mg N/L as nitrate, respectively (Fig. 3).

C. Spring 2025 PRB Performance Monitoring

Groundwater conductivity ranged from 196-474 µS/cm and average groundwater temperature was 11.6 °C. Groundwater nitrogen consisted of nitrate, ammonium, and organic nitrogen. Nitrite was below the LOQ in all samples. Similar to the fall and winter sampling events, there was higher nitrate at 20-25 ft compared to 10-15 ft below grade. The largest difference was observed at TCMW4 again where there was 7.3 mg N/L at 20-25 ft and 1.28 mg N/L at 10-15 ft. Upgradient wells at 10-15 ft and 20-25 ft below grade had 1.8 ±1.0 mg N/L and 4.7 ±1.9 mg N/L as nitrate, respectively. Downgradient wells at 10-15 ft and 20-25 ft below grade had 0.3 ±0.1 mg N/L and 2.0 ±1.6 mg N/L as nitrate, respectively (Fig. 3).

D. Summer 2025 PRB performance monitoring

Groundwater conductivity at the Tanbark Creek PRB site ranged from 200-630 µS/cm and groundwater temperature was an average of 16.5 °C. Groundwater nitrogen consisted of nitrate, ammonium, and organic nitrogen. Nitrite was always below the LOQ in all samples. Similar to all other seasons, there was higher nitrate at 20-25 ft compared to 10-15 ft below grade. The largest difference was observed at TCMW4 again where there was 7.1 mg N/L at 20-25 ft and 0.9 mg N/L at 10-15 ft. Upgradient wells at 10-15 ft and 20-25 ft below grade had 1.3 ±1.0 mg N/L and 4.4 ±1.9 mg N/L as nitrate, respectively. Downgradient wells at 10-15 ft and 20-25 ft below grade had 0.15 ±0 mg N/L and 2.0 ±1.4 mg N/L as nitrate, respectively (Fig. 3).

Downgradient of the PRB, monitoring well samples had higher alkalinity (76 ±21 mg/L) compared to upgradient monitoring wells (39 ±14 mg/L). These results are consistent with the denitrification reaction which produces bicarbonate ions and increases alkalinity. Summer 2025 pH levels ranged from 5.7 to 6.6. The optimal pH range for denitrification is 7 to 7.5 so values are below optimal conditions but still within the range that denitrification activity can occur (Saleh-Lakha et al. 2009). Dissolved oxygen concentrations were slightly higher in upgradient samples (1.7 ±0.7 mg/L) compared to downgradient samples (1.0 ±0.5 mg/L). All upgradient samples had positive ORP values while all but one downgradient sample had negative ORP values. Low oxygen values and negative ORP values are reflective of anoxic conditions which are required for denitrification to occur.

DOC was elevated in the downgradient samples (13 ±17 mg/L) compared to the upgradient samples. All but one upgradient sample was below detection (<1.00 mg/L). The presence of DOC in downgradient samples confirms that soybean oil emulsion which coats the sand grains within the PRB is metabolized by microbes into a dissolved form. The lack of DOC in the upgradient wells also confirms the soybean oil did not disperse beyond the original injection area.

Woodchip-based PRBs are known to initially release a pulse of DOC at similar concentrations measured at Tanbark and then return to steady-state conditions and last for decades (Robertson et al. 2008, Gilbert et al. 2019). Similarly, microbial degradation of vegetable oil emulsion to DOC supports heterotrophic denitrification. While vegetable oil PRBs for chlorinated solvent removal have continued to function for over 2.5 years (Borden and Lieberman 2009), there are less published studies on the lifetime of vegetable oil PRBs for nitrate removal. Less DOC was present in downgradient well samples in year 2 (13 ±17 mg/L) compared to year 1 (63 ±65 mg/L), possibly indicating a decline in the available carbon. However, the field parameters, alkalinity, DOC and nitrate data all indicate that denitrification occurred in the PRB in year 2.

Background levels of dissolved iron in the upgradient PRB monitoring wells were all below detection (<0.2 mg/L). Downgradient PRB monitoring wells had comparatively elevated iron of 7 ±10 mg/L. There was a slight decline in average iron concentration from 2024 (16 ± 14 mg/L) to 2025 (7 ±10 mg/L) suggesting that either sediment iron is depleted, or less carbon is available for microbially mediated dissimilatory iron reduction. Dissolved iron can reprecipitate into iron sulfide and iron oxide minerals when exposed to reduced sulfur compounds (H_2S, HS^-) and to oxygen, respectively. In fact, Borden and Lieberman 2009 found that elevated dissolved iron levels due to a vegetable oil based PRB reprecipitated within 100 ft downgradient of the barrier. The distance from the PRB where mineral precipitation occurs, also known as the redox recovery zone, is influenced by the natural groundwater sulfate concentration, groundwater travel time, rainfall, rate of mixing and other site-specific parameters.

E. Summer 2025 PRB carbon injection longevity monitoring

Surprisingly, soybean oil was not detected in any of the soil samples collected within the PRB zone (n=3). Although soybean oil itself was not detected in the soil within the PRB zone, DOC, a by-product of soybean oil was measured in water samples in the downgradient PRB monitoring wells. Soil total organic carbon (TOC) results from year 2 (Sept. 2025) were surprisingly different than results from year 1 (Nov. 2024) (Table 1). In year 1 the soil TOC data matched our hypothesis that sandy soils upgradient of the PRB (TCMWC1-25SB, TCMWC3-25SB, TCMWC4-25SB) would have less organic carbon than soil within the PRB zone (TCC2-

25SB, TCC4-25SB, TCC5-25SB). However, in year 2, the TOC content was similar or higher in soils upgradient of the PRB compared to samples within the PRB zone.

A possible explanation for this surprising result is that in year 1 samples were collected in the fall when trees had already lost their leaves and were no longer performing photosynthesis. In year 2, soil samples were collected in late summer when trees were undergoing photosynthesis. The PRB is situated in a woody area, and photosynthetically active trees may have been depositing organic carbon into the soil via root exudates. Tree root exudates are carbon-rich compounds that get transferred to soil via tree roots during photosynthesis. The upgradient monitoring well DOC samples were not consistent with the soil TOC near the upgradient monitoring wells which makes these results even more surprising but the soil TOC contains non-dissolved forms of carbon. In future monitoring years it will be useful to sample soil TOC in both summer and late fall to see if photosynthesis and root exudates mask the difference in carbon content between upgradient and within PRB soil samples.

Table 1: Soil TOC in µg carbon per gram (g) of soil. White rows represent samples collected near the upgradient PRB monitoring wells. Gray rows represent samples collected within the anticipated PRB zone.

TOC (µg/g)	Year 2 (Sept. 2025)	Year 1 (Nov. 2024)
TCMWC1-25SB	4820	<300
TCMWC3-25SB	3450	<300
TCMWC4-25SB	5000	<290
TCC2-25SB	2950	1850
TCC4-25SB	3960	7580
TCC5-25SB	5920	10400

F. Fall 2024 and Spring 2025 Tanbark Creek water quality monitoring

Overall, there were no unusual deviations in field parameters including dissolved oxygen, pH and ORP in porewater or surface water. All the porewater and surface water field data is within the range of what is considered “normal” and similar to what CCE and CLEAR have measured in other shallow creeks without PRBs. Thus, the pilot injection PRB is not negatively influencing nearby environmental conditions in the wetland or creek. To the best of our knowledge, we are the first group trying to directly link pilot PRB performance with offshore porewater/surface water quality and there have been lessons learned during this process.

In fall 2024 the strongest part of the Tanbark Creek SGD zone was within the first 10 ft from the shoreline which is consistent with measurements from previous years. The fall 2024 and spring 2025 24-hour average SGD rate at the mid-point of the SGD zone at station TCSP7_5 was 54 and 37 mL/min, respectively. At this station there was also a strong tidal trend with highest seepage rates measured at low tide and minimum seepage rate measured at high tide. In both seasons at high tide the seepage rate was still positive, which is indicative of strong groundwater drive and is one of the reasons this site was identified as suitable for a PRB.

The year 2 SGD rates were similar in magnitude to the daily average rate of 41 mL/min measured at the same location in 2021 before PRB installation. Thus, no change in SGD rate was observed since installation of the PRB. The sediment type at TCSP7_5 and TCSP7_25 remained consistent across both monitoring years. At 25 ft away from the shoreline at station TCSP7_25 the sediment was ~6 inches of muck which was starkly different than the sediment within the first 10 ft which consisted of sand and some gravel. Muck is much less conductive of groundwater. Consequently, the seepage data at TCSP7_25 was very noisy, and hovered around zero, with a 24-hour average of 9 and -0.7 mL/min in fall 2024 and spring 2025, respectively. At both stations and across both monitoring years, the average seepage rates collected in fall were higher than those collected in spring, likely due to higher water table causing a greater pressure gradient.

In fall 2024 and spring 2025 porewater nitrogen was primarily in the form of nitrate. In fall 2024, out of 17 nitrogen samples collected in a grid pattern throughout the cove, nitrate ranged from below detection (< 0.4 mg N/L) up to 5.44 mg N/L while TKN ranged from below detection (< 1 mg N/L) up to 2.5 mg N/L. In spring 2025, out of 13 samples collected throughout the cove, nitrate ranged from below detection (< 0.3 mg N/L) up to 4.64 mg N/L (Fig. 4) while TKN ranged from below detection (< 1 mg N/L) up to 4.22 mg N/L. In comparing the original porewater stations pre-PRB installation to post-PRB installation, the lowest average porewater nitrate was observed this year in spring 2025 (Fig. 5) despite slightly higher upgradient (untreated groundwater) nitrate (Fig. 3).

While expanding the offshore porewater stations into a grid throughout the cove was useful, so far it did not reveal any additional seepage zones than what was already known. The groundwater seepage seems to be occurring within the first 10 ft of the shoreline and our original porewater stations seem aptly placed to capture the flow and nitrogen concentrations. We will continue to monitor the expanded grid in year 3 to verify what we learned in year 2.

Overall, the SGD zone monitored in the cove downgradient of the pilot PRB within the first 10 ft is a focal point for groundwater discharge and is a mixture of groundwater that is not only coming through the PRB but from beneath, above, and around the PRB treatment area as well. The modeled saltwater interface beneath the injection wells is >100 ft below the top of the water table (Town of East Hampton Basis of Design Report Conceptual (30%) Design; Feb. 23, 2022; Memorandum pg. 4). Essentially, the fresh groundwater treated within the pilot PRB is approximately ≤ 20% of the total fresh groundwater column above the saltwater interface. Any nitrate present below or around the PRB could also be discharging into the cove and show up in the porewater samples. Furthermore, groundwater at this site contains TKN which is naturally converted to ammonium and subsequently to nitrate via nitrification in the presence of oxygen. Thus, any TKN present in groundwater downgradient of the PRB could be converted to nitrate and be present in the porewater. This year we confirmed there is enough time and oxygen present to re-oxidize reduced iron before it reaches the shoreline so it seems likely there is enough time and oxygen to convert reduced forms of nitrogen (TKN) to the more oxidized form (nitrate) as well.

The takeaway from the offshore porewater data is that the pilot PRB was initially intended to be bigger and it needs to be bigger to see a measurable decrease in porewater nitrate to provide more water quality benefits. It was designed as a proof of concept and is only treating a subsection of the aquifer. A much deeper and wider water column with nitrogen rich water is discharging into the cove. Based on the success of the current pilot, there is potential to expand the PRB deeper or further south where >20 mg N/L nitrate concentrations were present during site characterization. A groundwater tracer test can also help verify some of the original assumptions about groundwater direction in this region. If funding for PRB expansion becomes available, a tracer test will be included in future site characterization activities.

Lastly, in 2025 a green filamentous macro-algae bloom occurred during offshore porewater measurements. The algae hadn't been observed during previous measurements. However, the timing of the measurements may play a role in its observation and just because we hadn't seen it before does not mean it wasn't there. We plan to look for it and collect samples for algae identification in the year 3 monitoring period.

Figures:

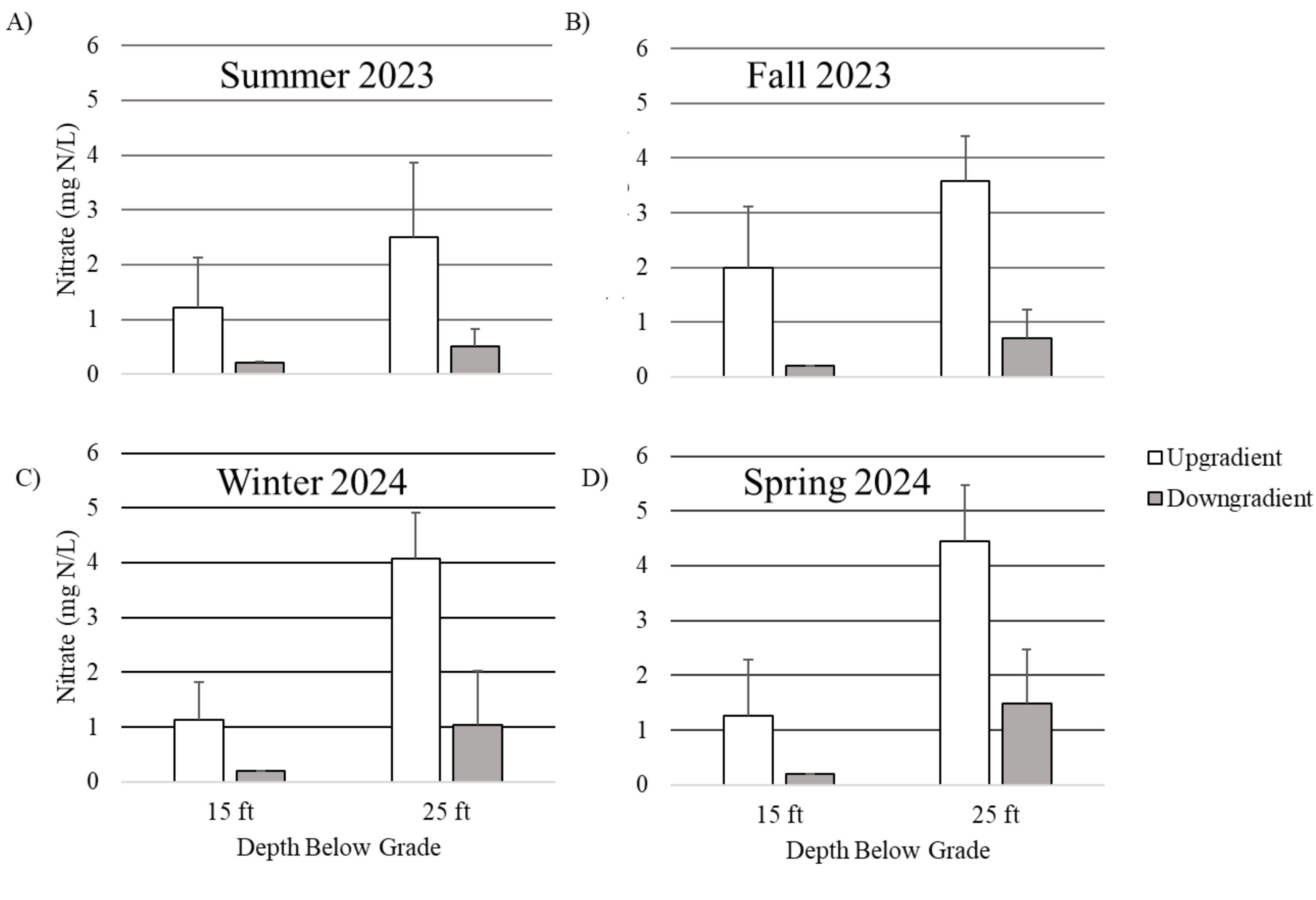


Figure 2: Year 1 of quarterly PRB performance monitoring for comparison with year 2. Average ± std. dev. of nitrate concentrations upgradient of the PRB (untreated groundwater) and downgradient (treated water) at 10-15 ft (n=4) and 20-25 ft (n=4) below grade.

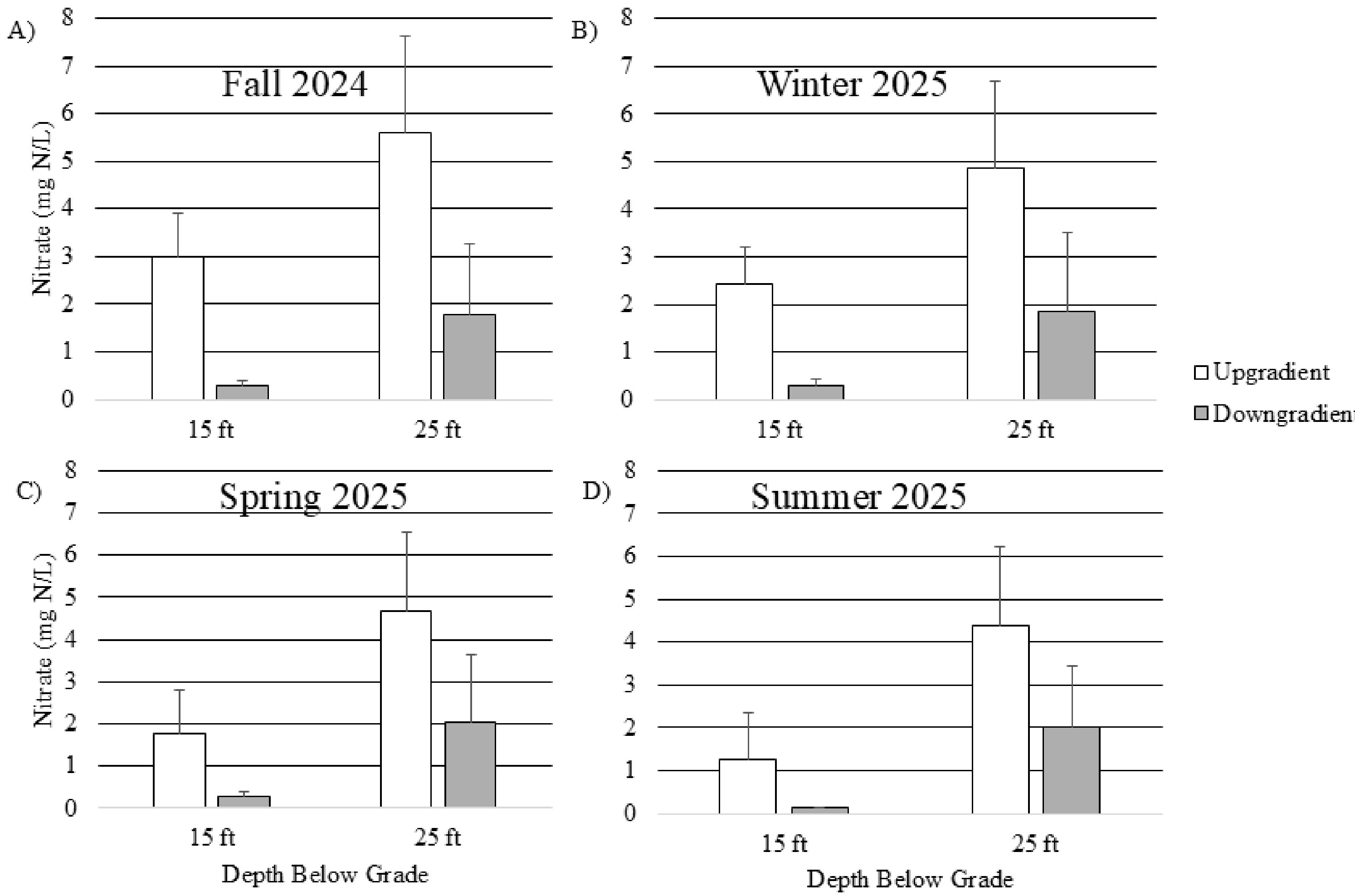


Figure 3: Year 2 of quarterly PRB performance monitoring. Average ± std. dev. of nitrate concentrations upgradient of the PRB (untreated groundwater) and downgradient (treated water) at 10-15 ft (n=4) and 20-25 ft (n=4) below grade. Please note the y-axis magnitude for year 2 is higher compared to year 1.

Figure 4: Porewater and surface water nitrate concentrations (mg N/L) in spring 2025. Expanded grid of porewater stations (red dots/yellow text), original 5 porewater stations (purple dots/yellow text) and surface water (red dots/blue text). Values below detection limit are indicated as BDL.

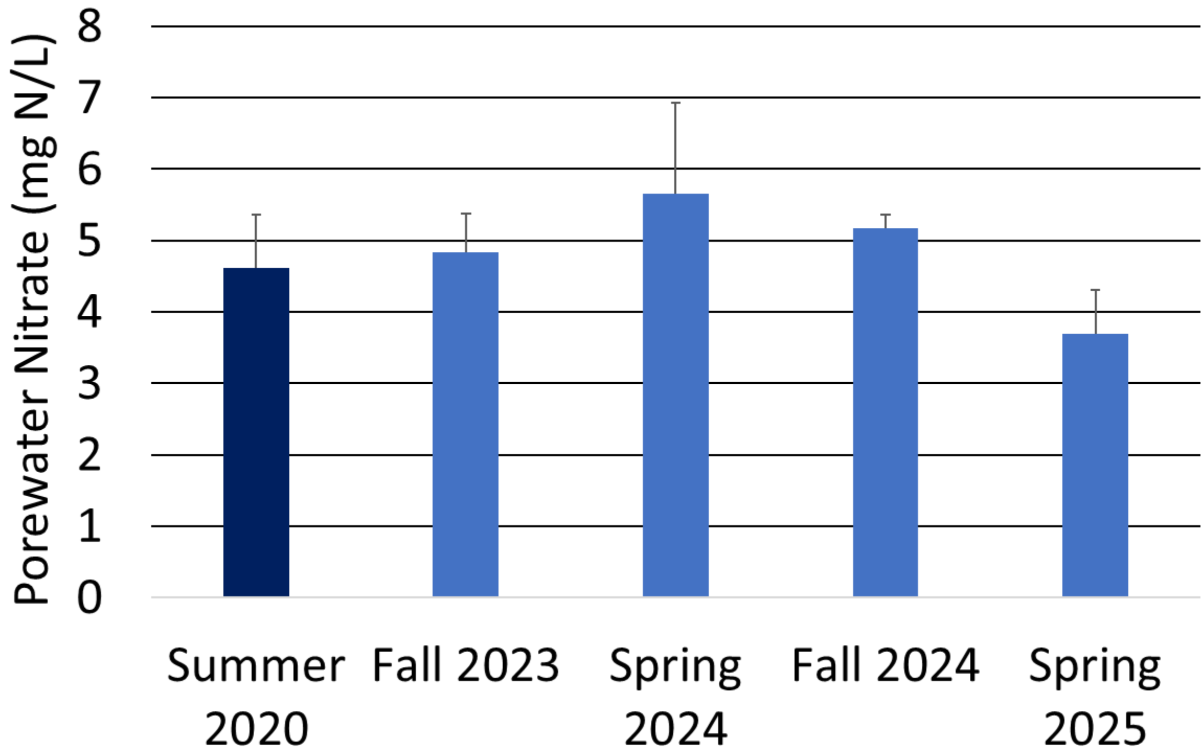


Figure 5: Comparison of original offshore porewater nitrate values (average ± std dev.) pre-PRB installation (summer 2020; n=3) to post-PRB installation Year 1 (fall 2023, n=5; spring 2024, n=5) and Year 2 (fall 2024, n=5; spring 2025, n=5).

References:

Borden, R.C., Lieberman, M.T. (2009). Passive Bioremediation of Perchlorate Using Emulsified Edible Oils. In: Stroo, H.F., Ward, C.H. (eds) *In Situ* Bioremediation of Perchlorate in Groundwater. SERDP/ESTCP Environmental Remediation Technology. Springer, New York, NY. https://doi-org.proxy.library.cornell.edu/10.1007/978-0-387-84921-8_8

Gilbert, O., Assal, A., Devlin, H., Elliot, T., & Kalin, R. M. (2019). Performance of a field-scale biological permeable reactive barrier for in-situ remediation of nitrate-contaminated groundwater. *Science of the total environment*, *659*, 211-220.

Town of East Hampton Basis of Design Report Conceptual (30%) Design. February 28, 2022. CDM Smith

Robertson, W. D., Vogan, J. L., & Lombardo, P. S. (2008). Nitrate removal rates in a 15-year-old permeable reactive barrier treating septic system nitrate. *Groundwater Monitoring & Remediation*, *28*(3), 65-72.

Saleh-Lakha, S., Shannon, K. E., Henderson, S. L., Goyer, C., Trevors, J. T., Zebarth, B. J., & Burton, D. L. (2009). Effect of pH and temperature on denitrification gene expression and activity in Pseudomonas mandelii. *Applied and environmental microbiology*, *75*(12), 3903-3911.

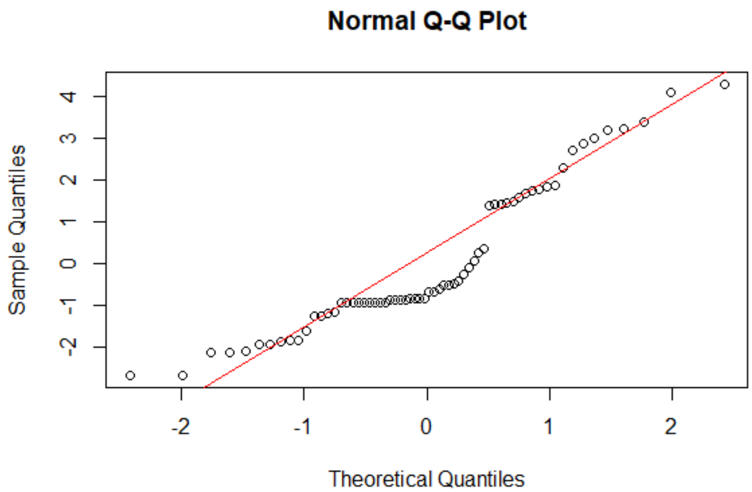
Exhibit A Table 1: Year 2 nitrate concentrations analyzed in R using a Shapiro-Wilk normality test and Wilcoxon rank sum tests. Eight values for each season are from the 8 monitoring wells in each group (2 wells at each of 4 cluster wells). Values below detection are considered ½ of the detection limit (i.e. <0.4 = 0.2).

Group	Season	Nitrate_Value
Upgradient	Fall	1.62
Upgradient	Fall	2.19
Upgradient	Fall	3.04
Upgradient	Fall	6.12
Upgradient	Fall	4.18
Upgradient	Fall	7.51
Upgradient	Fall	3.12
Upgradient	Fall	6.56
Upgradient	Winter	1.82
Upgradient	Winter	2.04
Upgradient	Winter	3.71
Upgradient	Winter	5.02
Upgradient	Winter	1.82
Upgradient	Winter	5.40
Upgradient	Winter	2.39
Upgradient	Winter	7.03
Upgradient	Spring	1.10
Upgradient	Spring	2.02
Upgradient	Spring	3.57
Upgradient	Spring	4.64
Upgradient	Spring	1.09
Upgradient	Spring	4.79
Upgradient	Spring	1.28
Upgradient	Spring	7.30
Upgradient	Summer	0.15
Upgradient	Summer	1.88
Upgradient	Summer	3.07
Upgradient	Summer	4.26
Upgradient	Summer	0.96
Upgradient	Summer	4.23
Upgradient	Summer	0.90
Upgradient	Summer	7.10
Downgradient	Fall	0.20
Downgradient	Fall	0.54
Downgradient	Fall	0.20
Downgradient	Fall	3.72
Downgradient	Fall	0.20
Downgradient	Fall	0.20

Downgradient	Fall	0.50
Downgradient	Fall	2.69
Downgradient	Winter	0.20
Downgradient	Winter	0.40
Downgradient	Winter	0.20
Downgradient	Winter	4.25
Downgradient	Winter	0.20
Downgradient	Winter	0.20
Downgradient	Winter	0.55
Downgradient	Winter	2.56
Downgradient	Spring	0.20
Downgradient	Spring	0.73
Downgradient	Spring	0.20
Downgradient	Spring	4.14
Downgradient	Spring	0.20
Downgradient	Spring	0.20
Downgradient	Spring	0.48
Downgradient	Spring	3.02
Downgradient	Summer	0.15
Downgradient	Summer	0.82
Downgradient	Summer	0.15
Downgradient	Summer	3.94
Downgradient	Summer	0.15
Downgradient	Summer	0.48
Downgradient	Summer	0.15
Downgradient	Summer	2.87

Results of the Shapiro-Wilk normality test:

W = 0.90529, p-value = 0.0001274



Due to significant p-value in the Shapiro-Wilk normality test and due to s-shaped Q-Q plot, the assumption of normally distributed residuals was not met. Therefore, a repeated measures analysis of variance can't be used because the test assumes normally distributed residuals. Therefore, differences in nitrate concentrations between upgradient and downgradient wells were assessed for each season using Wilcoxon rank-sum tests. Because the data contained tied values, p-values were calculated using a normal approximation to the Wilcoxon rank-sum distribution. All p-values were less than 0.05 indicating that nitrate concentrations are significantly lower at downgradient wells compared to upgradient wells across all seasons.

Exhibit A Table 2: Results of Wilcoxon rank-sum tests

Season	W	p-value	Interpretation
Fall	6	0.006977	Significant difference
Winter	9	0.0172	Significant difference
Spring	9	0.01729	Significant difference
Summer	13	0.0486	Significant difference