

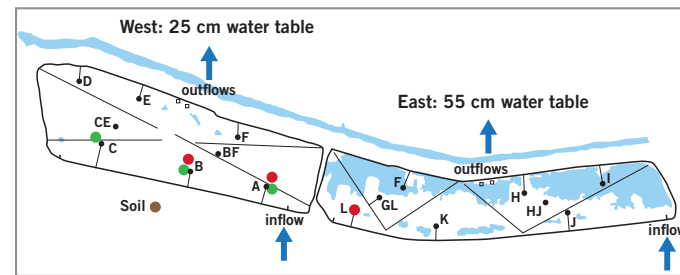
Findings

In Phase I of this project (2011–2013), we sampled the 15-year-old restored wetland on Twitchell Island (121.65°W, 38.11°N) in the SF Delta region in February and August of 2011 to determine the microbial contribution to carbon sequestration and emission from a restored wetland.

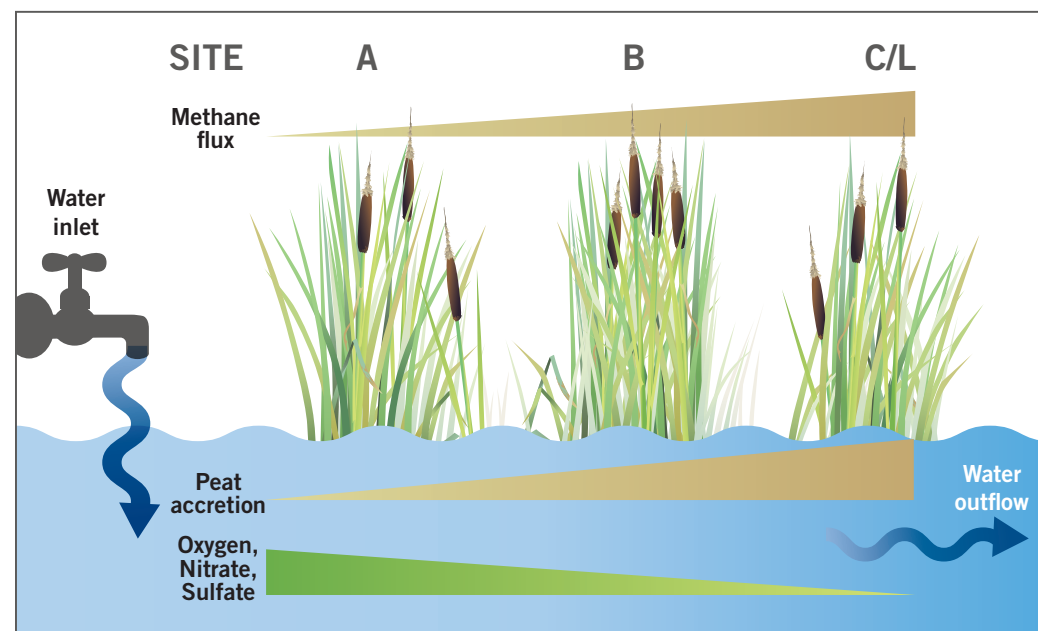
In collaboration with the California Department of Water Resources (DWR) and the U.S. Geological Survey (USGS), we sampled microbial communities in bulk soils and plant rhizospheres and measured carbon dioxide and methane production. DNA sequencing results demonstrated a clear relationship between microbial community composition and geochemical gradients. Oxygen was generally low, and we saw a decrease in sulfate-reducing bacteria and an increase in methanogenic archaea with decreasing sulfate availability and increasing methane production. Microbial community composition clustered according to plant type (tule rhizome, cattail rhizome, bulk soil) and sampling location (sites A, B, C/L). This project successfully demonstrated the ability to relate microbial community profiles with geochemical gradients, using DNA sequencing to link species abundances to greenhouse gas flux.

In Phase II of this project (2013–present) we sampled a suite of wetlands throughout San Pablo Bay, Suisun Bay, and the Delta regions of SF Bay, encompassing a salinity gradient from freshwater to marine. Covering five historic and six restored wetlands, we sampled wetland soils in a variety of plant communities. Our preliminary results revealed a difference between historic and restored wetland soil methane

production, with historic wetlands producing much less methane on average than restored wetlands. This disparity may be the result of larger labile carbon pools in younger wetlands. As expected, freshwater wetlands were responsible for higher methane production rates than more saline sites where sulfate availability diminished methane production. Our future DNA sequencing will uncover the composition of the associated microbial communities along this salinity gradient and will reveal if variations in methane production were a result of differences in microbial community composition, microbial metabolic rates, or a combination of the two.



Overview of Twitchell Island sampling locations. West and east sections of the pond had artificially maintained depths of 25cm and 55cm, respectively.



Schematic of sampling site locations A, B, and C/L along oxygen and nutrient gradients.

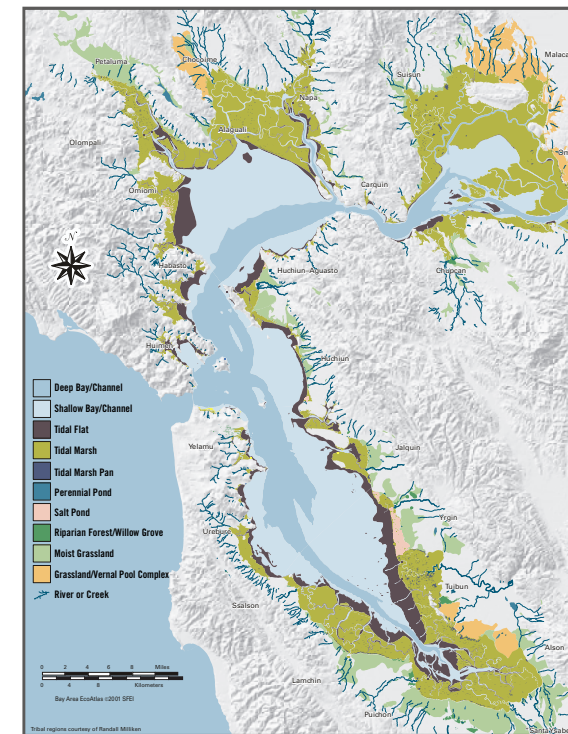
Restoring Our Wetlands: The Microbial Balancing Act

Wetland Restoration in San Francisco Bay

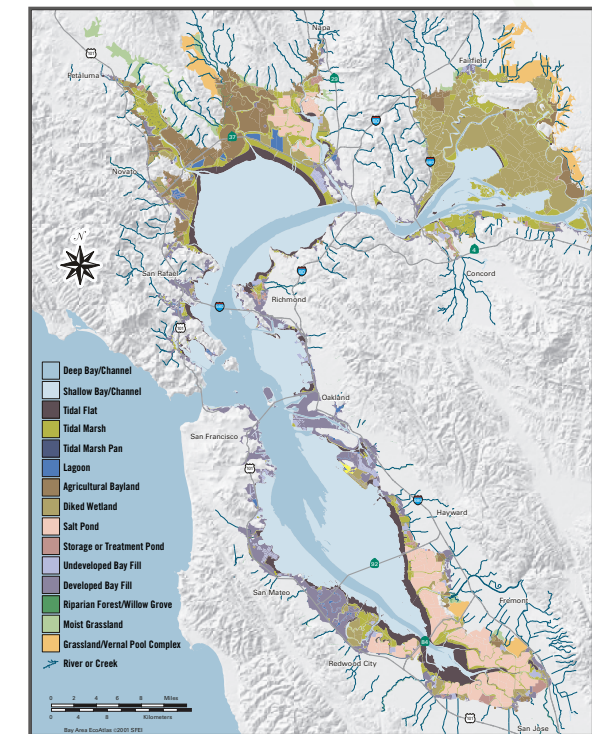
Wetlands have been described as “nature’s kidneys,” responsible for filtering nutrients and protecting inland habitats from tidal action. Wetlands also have the potential to serve as carbon sinks and to sequester atmospheric carbon as the result of high plant productivity and low decomposition rates.¹

In the past 150 years, San Francisco Bay (SF Bay) lost 80–90% of its coastal wetlands² due to industrial development and agricultural conversion. Years of agricultural and salt pond farming have resulted in significant land subsidence, levee stress, and flood risk throughout SF Bay. To mitigate these threats, multiple restoration projects have reopened these coastal habitats to tidal action. These projects aim to restore native plant and animal species, reverse land

subsidence, and earn carbon sequestration credits as a result of long-term carbon burial. A pilot project on Twitchell Island in the Sacramento-San Joaquin River Delta was established in 1997 to measure rates of carbon flux and carbon accrual in a freshwater wetland. This project demonstrated successful accretion rates of 2 inches/year³ and has provided valuable information on seasonal variations in greenhouse gas flux from a restored wetland.



Historical (circa 1800) baylands



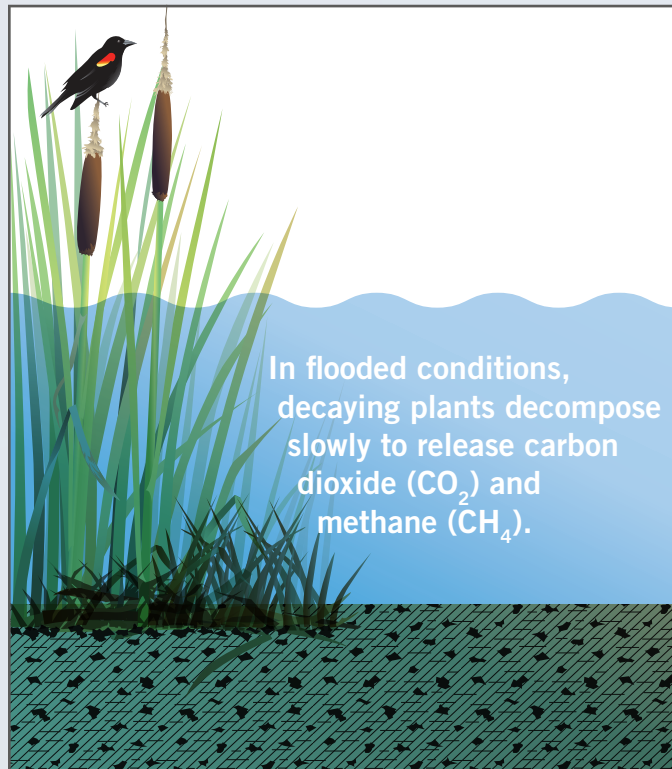
Modern (circa 1998) baylands

¹Connor et al., 2001; Hussein et al., 2004.

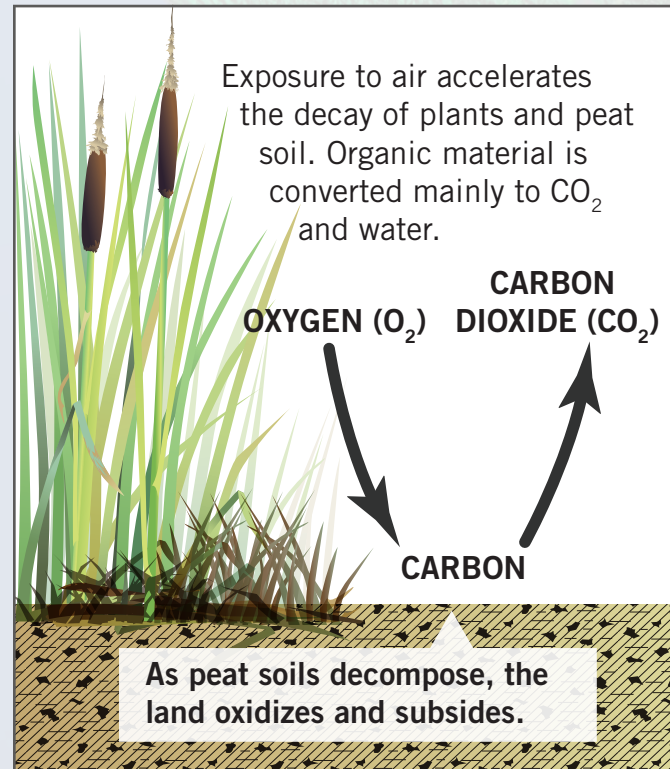
²Williams and Faber, 2001; H.T. Harvey and Associates, 2008.

³Miller et al., 2008.

Anaerobic Conditions: Oxygen Poor



Aerobic Conditions: Oxygen Rich



Schematic of anaerobic conditions that promote methane production, and aerobic conditions that promote carbon dioxide production in wetlands.

Wetlands and Microbes

All wetlands are capable of sequestering and storing carbon through photosynthesis and the accumulation of organic matter in soils, sediments, and plant biomass. Waterlogging of wetland soils limits oxygen diffusion into sediment profiles, creating anaerobic conditions.

These conditions slow decomposition rates, leading to the buildup and storage of large amounts of organic carbon in wetland sediments. However, anaerobic conditions also promote the production of greenhouse gases, offsetting the positive effects of carbon accrual. The balance between wetlands serving as a greenhouse gas source or sink is dictated by the aboveground plant species' primary productivity and the metabolic activity of belowground microbial communities. These belowground microbial communities are responsible for the production and consumption of greenhouse

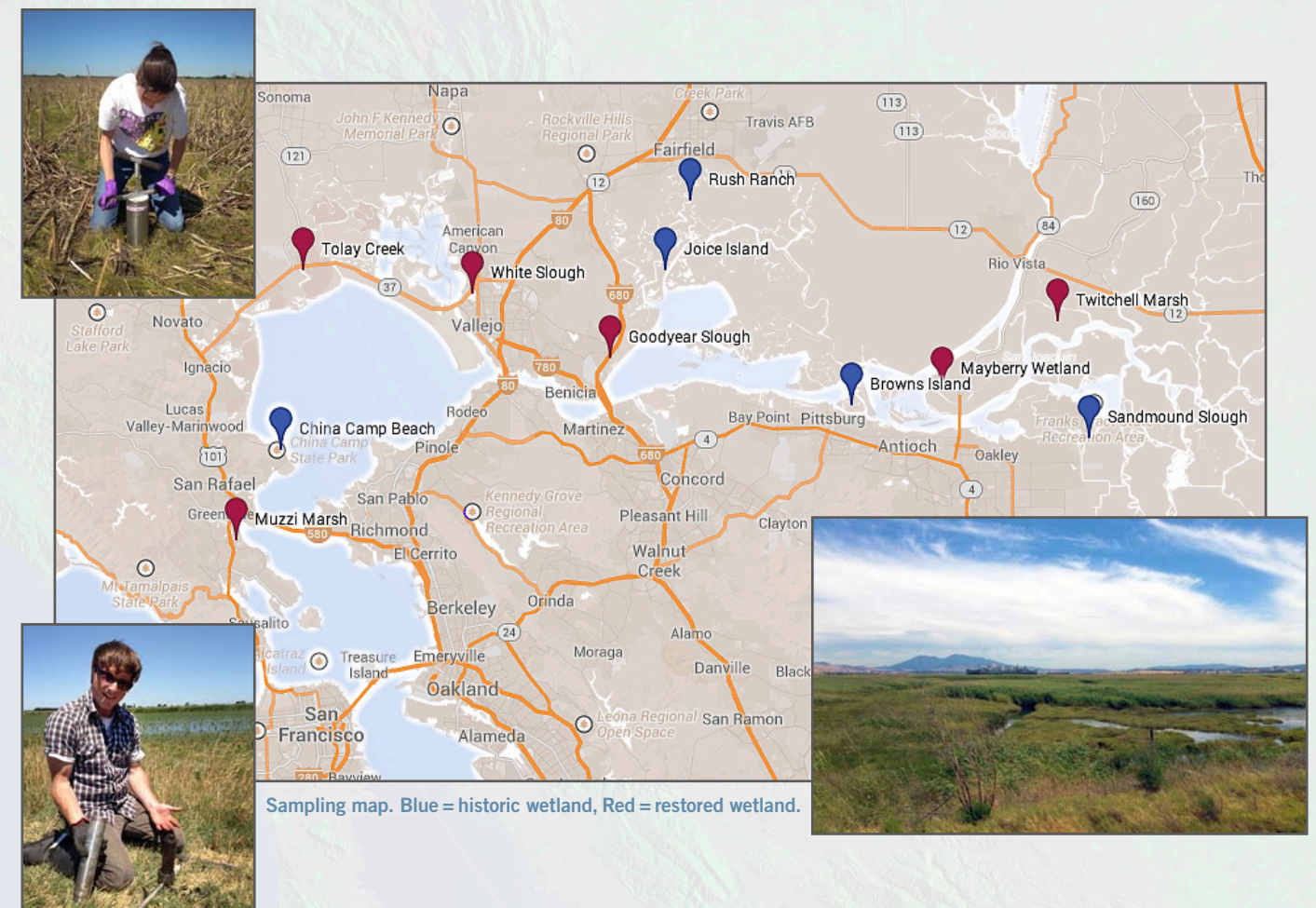
gases such as carbon dioxide (CO₂), methane (CH₄), nitrous oxide (N₂O), and sulfur dioxide (SO₂). An improved understanding of microbial community dynamics in natural and restored wetlands will help guide future wetland engineering to promote optimal carbon sequestration conditions. Our project focuses on characterizing the microbial communities along a spectrum of historic and restored wetlands, as well as salinity regimes, to determine microbial community dynamics and the wetland's ability to provide a viable long-term carbon-storage solution.

Sampling San Francisco Bay Region Wetlands

Sample collection consisted of taking soil cores for DNA, RNA, and single-cell sequencing as well as soil-pore water for geochemical analyses. Soil samples were extracted for total DNA and RNA. High-throughput DNA sequencing targeted the 16S ribosomal RNA (rRNA) gene to profile the bacterial and archaeal populations.

Metagenomic (DNA) sequencing allowed for profiling the metabolic potential of the microbial population, and metatranscriptomic (RNA) sequencing revealed the active metabolic pathways. Single-cell genome sequencing enabled the reconstruction of metabolic and enzymatic pathways in novel organisms. All sequencing was performed on the Illumina HiSeq and MiSeq platforms at the DOE Joint Genome Institute (JGI). Sequencing analyses were performed at the JGI in collaboration with the resources at the DOE National Energy Research Scientific Computing Center (NERSC).

Greenhouse gas flux from wetland soils was measured through a variety of methods. For the February 2011 sampling at the Twitchell Island wetland, soil subsamples were incubated in a laboratory for two weeks to evaluate CO₂ and CH₄ production. For the August 2011 sampling, stationary whole plant chambers were deployed to measure both belowground microbial activity and aboveground primary productivity. For the 2013 sampling of the SF Bay wetlands, intact soil cores were collected adjacent to the DNA/RNA core sample. Intact soil cores were measured for CO₂ and CH₄ production using a Los Gatos Research Greenhouse Gas Analyzer.



Sampling map. Blue = historic wetland, Red = restored wetland.