Data Collection for Oyster Mariculture Interactions with Seagrass

CMP Final Report

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COASTAL MANAGEMENT PROGRAM –CYCLE 26

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EXECUTIVE SUMMARY

This project set out to measure potential sediment generation at cultivated oyster mariculture (COM) operations in TX, then apply that knowledge to simulate sediment plumes with similar characteristics at test seagrass beds, to measure if there were impacts to the seagrass. . It should be noted that “sediment plumes” or “sediments” are used here in the general sense of suspended particles of inorganic or biotic nature that can settle or attenuate light penetration into the water column, regardless of source (NOAA). Observations and measurements made at shallow water research COM study sites (those accessible by walking), and a deep water commercial scale suspended cage COM farm (“deep” referring to unwalkable and requiring boat access: 6 – 10 ft depth in this case), characterized sediment disturbances with respect to light attenuation, and suspended and settleable solids. Aerial imagery and analyses tracked the sediment plume trajectory and persistence. Shallow COM operations with persons walking had the potential to produce heavier sediment plumes that could travel 100 ft. In contrast, measurements at a deep water COM farm where boat-based operations leave bottom sediments undisturbed, produced smaller, shorter lived sediment plumes that originated only from the suspended cages and proved difficult to measure. The former sediment plumes appeared to be largely lithogenous, whereas the latter appeared mostly biogenous. However, the study was unable to measure and observe COM disturbances in all possible operational activities, or weather and current conditions, so the plume metrics produced here cannot be taken as absolute representative limits, but rather should be treated as observations indicative of the range of values observed for the experiments performed here.

At a seagrass study site, sediment disturbances were created by persons walking along a disturbance baseline for standardized times (person-minutes; p-m). Sediment plume characteristics measured here were largely intermediate between those of the two types of COM operations. These plumes were observed to travel at widely varying rates ranging from over 300 ft at ~10 ft/min with strong current, to almost not at all, or somewhere in between. Plume travel in the very shallow seagrass beds of this study was dominated by tidal hydrology, with only a relatively small contribution from wind, and sometimes sediments actually moved in a counter-current fashion to the wind. Large tanker shipping through the Aransas Pass channel 5 miles to the South, could quickly reverse and dominate sediment plume movements as well as seagrass leaf orientation.

The seagrass sediment disturbances were applied according to an experimental design, and subsequent adaptations to unexpected hydrological challenges, that resulted in Tracks and Zones within the Tracks, that differed by intensity and duration of sediment plumes. In the Zones nearest to the disturbances (A), Track 3 (Heavy) accumulated 811 p-m of disturbance over 5 months, while Track 2 (Light) accumulated 455 p-m of disturbance over the same period. Track 1 and an up-current zone in each Track served as undisturbed controls. The seagrass

indicators of areal seagrass bed coverage, leaf density, leaf length, biomass, belowground to aboveground biomass ratio, epiphyte to seagrass biomass ratio, red to green epiphyte fluorescence ratio and image-based epiphyte accumulation %, were measured at different distances from the sediment plume source (50, 100 or 200 ft for Zones A – C respectively) along each of the 3 disturbance Tracks (treatments). Seagrass and invertebrate sampling occurred prior to (Pre), during (Mid) and following (Post) sediment disturbance applications. The BACI experimental design allowed comparisons between disturbance intensities (treatment Tracks), at increasing distance from each disturbance (Zones A – C) and across time (Pre to Post).

Except for the invertebrate sampling, seagrass indicators exhibited no significant differences between disturbance intensity treatments or with distance from the disturbance across all times sampled. The expected seasonality of seagrass indicators (eg reduced leaf length and biomass in November) were observed similarly in all comparisons. For the invertebrates, there was a marginally significant variation in community dissimilarity at different distances from disturbance as well as a decreasing abundance of invertebrates. Distance along each track coincided with increasing depth (8 - 11 cm depth difference along each track).

The overarching conclusion from this short-term experimental work is that we found no evidence to suggest that an unwalkable deep water suspended cage COM operation, as currently permitted with *minimum* 200 ft clearance from seagrass, results in sediment or light attenuation levels that would be likely to impair seagrasses (*Halodule wrightii* specifically) over the short term. This applies to suspended cage COM where there is little disturbance of the bottom. There is less certainty about any potential impacts from shallow COM operations as they tend to have greater sediment plume levels due to bottom disturbance. Importantly, as a short term experiment, the results do not necessarily apply to any potential long-term impacts.

Another potential limitation in application of these conclusions is the difference in composition of the sediment plumes at COM sites (shallow vs deep) and those generated at the seagrass study site. Plumes from the latter presumably derived from epiphytes and detritus. In contrast, sediments from shallow COM sites in this study derived mostly from disturbed bottom sediments which would be a combination of native sediments plus any accumulated biodeposits which might provide a different nutrient profile compared to that from seagrass sediment plumes.

The indispensability of aerial imagery to this project should be noted as it provided unique visualization and analysis of sediment plumes. Hydrologic complications made measurements from fixed stations difficult to interpret or accept as representative in the face of shifting currents. To continue to fill important knowledge gaps in the future, longer term monitoring making use of Acoustic Doppler Current Profilers (ADCP) would provide a spatial understanding of hydrology and aid understanding of sediment accumulation patterns. Lastly, another knowledge gap to consider is understanding the potential for impact from *in situ* cage cleaning. The potential economic benefit is clear, but little is known about what, if any, impacts might occur.

INTRODUCTION

Overview

Cultivated Oyster Mariculture (COM) is a relatively new and growing industry in Texas. At the time this project initiated, there were only two newly operating COM farms in the state, and as of project end, there are 8 permitted operations. The detailed permitting process includes requirements for siting to assure protection for the state's seagrass resources. These requirements include that COM operations must be off-bottom, non-dredging and located a minimum of 200 ft from any seagrasses. Routes of egress must also be approved to avoid disruption of seagrass. Though oysters and seagrass commonly co-occur in nature, there is little information available regarding the efficacy of the size of this buffer zone, or about potential interactions between suspended cage COM and seagrasses in Texas. There are concerns that turbidity or suspended sediments from the oyster farms could interact with nearby seagrasses and, over time, cause negative impacts. Oysters expel biodeposits of feces and pseudofeces, and the mariculture hardware cages and bags accumulate biofouling communities which can be released upon handling in the course of normal operations. It can be attractive to locate the farm in shallow waters near-shore because the farm could be operated without boats. In such cases, there would be repeated wading to and around the farm which stirs up the sediments. Suspended particulate matter, of organic or inorganic matter (respectively biogenous or lithogenous) can negatively impact seagrass by attenuating the light reaching seagrass, settling and burying the seagrass in excessive quantity, or by altering the nutrient environment of the seagrass. Oysters can have positive impacts on seagrasses as well, in either natural or COM situations, by filtering phytoplankton from the water, reducing wave energy and by increasing nutrients in the seagrass sediments.

To begin to address the knowledge gaps, Texas A&M University – Corpus Christi (TAMUCC) used CMP Cycle 26 funds to assess sediment plumes that might be generated by COM operations as well as the effects of different plume characteristics on seagrasses to help inform coastal managers in COM siting decisions. It should be noted that "sediment plumes" or "sediments" are used here in the general sense of suspended particles of inorganic or biotic nature that can settle or attenuate light penetration into the water column, regardless of source (NOAA). TAMUCC simulated the effect of COM sedimentation near seagrass beds with a goal to test the efficacy of different buffer distances in sand- and mud-dominated sediments. Sediment plume trajectories, intensities and persistence were observed and measured at existing research and commercial COM sites to develop a sedimentation model that estimated plume characteristics to guide application at seagrass study sites. These results were then used to develop standard disturbance protocols that were applied repeatedly at seagrass study sites where abiotic and biotic indicators were monitored for cumulative sedimentation effects. Analysis of project data established sediment plume parameters at COM and SG sites, and that short term application of such sediment disturbance plumes at seagrass beds did not have any statistically significant negative impacts on seagrass over the short term. Measured sediment plumes produced preliminary data on sediment plume characteristics at various distances from their source, and how these sediment plumes differed based on the type of COM operation or seagrass bed that served as their source. This expanded the knowledge base regarding the efficacy of buffer zone size on SG protection. The project's outcomes include data on the relationships between disturbance activity, sediment transport/plume characteristics, and seagrass status.

The data collected by this project will inform resource managers and stakeholders in their efforts to develop a robust sustainable oyster mariculture industry while affording maximum protection of seagrass resources. The project will provide critical data on the quantitative relationships of potential COM-seagrass interactions and contribute to the beneficial economic impacts derived from productive COM that promotes seagrass sustenance of other marine resources.

Oysters

Oysters are filter-feeders and as such they filter sediments, detritus, small phytoplankton, particulate-bound nitrogen and phosphorus, and other particulate organic matter from estuarine waters, which increases water clarity (Ward et al. 2004; Bayne et al. 1983; Newell et al. 1996; Peterson & Lipcius 2003; Plutchak et al. 2010). When oyster filtration is added to small-scale ecosystem models and large-scale hydrodynamic models, results include clearer water, deeper light penetration, and greater light availability to submerged aquatic vegetation (North et al. 2010; Cerco et al. 2007). Oysters also release feces and pseudofeces that sink to the ocean floor, labeled as biodeposits (Shumway et al. 1985; Beninger et al. 1999). Natural oyster beds typically have enriched levels of sediment porewater nutrients, which can be beneficial for seagrasses (Booth and Heck 2009). Mariculture hardware such as cages and bags can also accumulate biofouling communities which can be released upon handling during normal operations.

Seagrasses

Seagrasses are important ecosystem engineers and provide important ecosystem services, such as habitat provision (Heck et al. 2003); sediment stabilization (Christianen et al. 2013); wave attenuation (Fonseca & Cahalan 1992); carbon sinks (Duarte et al. 2013); and neighboring system subsidization (Heck et al. 2008). Their primary productivity requires abundant light and clear water to permit light penetration to the bottom. It is essential to maintain conditions that facilitate seagrass productivity and to understand how the health of seagrass is affected by a changing environment. Seagrasses are declining worldwide at a rate of 7% per year since 1990 (Waycott et al., 2009). There are many factors contributing to this decline, but one such pressure is aquaculture structures located in close proximity to seagrasses, especially if they are poorly managed (Turschwell et al. 2021; Murata et al. 2021). Due to their need for adequate irradiance to support photosynthesis, seagrass beds are confined to shallow waters where their survival within the intertidal zone is limited by the tolerance of desiccation (Koch 2001) and/or wave exposure (Koch et al. 2006). These are also preferred locations for many types of aquaculture facilities, which sets up the potential for conflict and is the reason for careful permitting considerations in Texas.

Potential Interactions Between Cultivated Oyster Mariculture and Seagrasses

Due to the potential for seagrasses and some COM grow-out sites to be located in similar areas, its important to understand the effects that they might have on each other. Aquaculture can cause eutrophication in the coastal environment (Grant et al. 2005) if the biodeposition caused by cultured organisms exceeds their removal through decomposition and by water exchange (Murata et al. 2021). Biodeposits transfer nutrients from the water column to the seabed and this can increase the nutrients and organic content of sediment under these farms and in some cases enhance seagrass growth (Crawford et al. 2003, Dumbauld et al. 2009). Excess nutrients can also lead to increased algal epiphyte coverage of seagrass leaves (Huang et al 2023; Allen & Williams. 2003; Borum 1987) which can negatively affect seagrass metrics such as biomass, shoot density, photosynthetic rate, and leaf growth rate (Dimech et al., 2000; Sand-Jensen 1977; Nelson 2017). Mussel aquaculture led to a loss in percent areal coverage by seagrass (Neckles et al. 2005). Multiple studies showed physical damage to seagrass due to husbandry activities such as farm access by wading, placement of aquaculture structures over or in seagrass beds (Everett et al. 1995) or harvesting from the bottom with seagrass (Tallis et al. 2009; Cabaco et al. 2005). The Tallis study found different degrees of reduced productivity, leaf length and density for eelgrass, depending on the culture method. They compared longlines (off-bottom), dredging of bottom culture and hand-picking from bottom culture. All had impacts but the dredging produced large decreases (70%) in density and production. They found direct effects of oyster harvesting from either hand picking or dredging (benthic practices) reduced the density of eelgrass due to breakage of shoots or rhizomes. The longline had the least impact, but impacts were oyster density-dependent (Tallis et al. 2009). Other aquaculture activities impacting the benthic environment outside of Texas include dumping of shells (Orth et al. 2012), mechanical raking (Peterson et al. 1987), or even trampling (Short et al. 2011) producing concerns for seagrass growth. Skinner et al. (2013) showed that impact of suspended bag oyster aquaculture (SBOA) directly over seagrass had negative effects on *Zostera marina* in the Atlantic Canadian Bays, seen as a 57% decline in above-ground biomass over 3 years. They hypothesized that shading from aquaculture equipment reduced productivity, reproductive viability, and survival. Avoidance or careful management practices can reduce impacts to seagrasses (Tallis et al 2009). It is reiterated that Texas does not allow direct contact of COM activities with seagrasses, nor are any on-bottom farming practices allowed. For example, floating bags, which are being used in Texas, have fewer environmental effects than on-bottom techniques (NSW DPI 2021).

A minimum buffer zone away from seagrass is a straightforward approach to protect seagrasses. An important question is what is a safe distance for seagrass beds to not be negatively impacted by oyster farms? Texas has followed other states in implementing a 200 ft minimum distance from any seagrasses. However, there are few studies that investigated impacts of off-bottom oyster mariculture with respect to indirect effects at some distance. Rubino and Fox (2021) studied potential impacts of off-bottom longline culture at a shallow wadable research COM in Copano Bay, TX. This site was surrounded by low density *Halodule wrightii*. A variety of seagrass parameters were measured over a growing season, but they found no evidence of significant impacts. An important target of their study was suspended sediments that were quantified by the amount settling into sediment traps. Fine suspended particles can travel in currents and later settle into seagrass beds.

Seagrass-Sediment Interactions

Natural events, such as storms and runoff can transport sediments into estuaries. This is exacerbated by Human based activities, such as agriculture, mining, road construction (Kirkman et al. 1990; Fourqurean et al. 2004; Suykerbuyk et al. 2015; Saunders et al. 2017). Suspended sediments reduce light available to seagrass which affects productivity (Duarte et al. 1993; Dennison et al. 1993). This is a major driver of seagrass losses (Waycott et al 2009).

Acute and chronic activities that increase fine sediment concentrations, can result in complete and perhaps irreversible, extinction of seagrass meadows (Short et al. 1996; Erftemeijer & Lewis 2006; Cabaco et al. 2008; Maeztu et al. 2020). Suspended sediment present in the water column attenuates sunlight, which reduces the amount of available light for seagrass. Seagrass only thrives in areas where there is sufficient light reaching the canopy so suspended sediments reduce colonizable benthic habitat. Cabaco et al. (2008) studied a wide range of burial and erosion disturbances to see if impacts to different seagrass species were related to plant size. Burial thresholds were analyzed in order to help identify seagrass characteristics to better predict burial effects. Sediment burial affected seagrass growth and survival negatively, with large species being less susceptible to burial than smaller fast-growing species. In another study done by Maeztu et al. 2020 investigated potential causes of seagrass decline, by comparing light climate and substrate physical and chemical characteristics at habitats with and without seagrass. They found light not to be an issue for their intertidal seagrass, but the fine sediment did exert stress by deoxygenation of the root zone. They highlighted the critical importance of catchment strategies that reduce fine sediments in estuaries. Erftemeijer et al. (2006), reviewed 45 different case studies regarding channel dredging near or around seagrass beds. Channel dredging and disposal of dredged material led to a temporary decrease in water transparency. For over half of the case studies there was a loss of around 21,0123 ha of seagrass due to dredging, primarily due lowering light penetration to seagrass, producing sub lethal effects or even death.

Project Justification and Approach

There are knowledge gaps in how an off-bottom mariculture facility might impact seagrass at some distance away, since there is no direct shading or operator contact if they have a clear path of egress. Some questions that arise include: 1) what might travel from the COM farm to a seagrass bed 200 ft away? Suspended fine sediments that might bury seagrasses? Nutrients that might result in eutrophication? Suspended particles that might increase turbidity and result in shading of the light required by seagrasses ? Algal propagules or other biota that colonize the mariculture hardware? Answering these questions will require knowledge of what moves away from COM sites, and how far and how fast and how much. Then it also requires knowledge of how these things might actually impact seagrass. Since currently permitted COM operations have been sited to avoid potential conflicts with seagrasses, there is no one place where a study can be performed. The approach taken in this project was to perform studies at COM sites to understand what suspended materials (hereafter referred to in a general sense as "sediments" because they can attenuate light penetration and settle) travel away from the sight. This includes characterization of light attenuation and % organic composition, and how far and how fast they travel. Then, the second part of this study tries to find out how much suspended "sediments" might impact seagrasses by creating simulated sediment plumes at seagrass beds and studying impacts to the seagrass bed characteristics.

Goals

Texas Cultivated Oyster Mariculture (COM) may impact seagrasses (SG). Oysters and SG commonly co-occur in nature, but few SG studies exist for suspended cage culture. A critical issue is the buffer distance needed to protect SG beds from COM sedimentation. Suspended sediments reduce light to SG, but the relationships between COM operations, suspended sediments and SG are not known. COM siting is based on the untested supposition a 200 ft buffer protects SG. We will assess sediment plumes generated by COM, and the effects of different plume characteristics on SG, for data to inform coastal managers in siting decisions.

Study of an operating COM near seagrass would be ideal for a study, but permitted operations were conservatively sited and not suitable to evaluate a range of distances. Adding a pilot scale COM to a SG study is beyond budget and timeline limitations. Instead, we devised a pilot scale simulation of the sedimentation effects of COM, near SG beds, to determine best buffer distances in sand- and mud-dominated sediments. Innovative tools will characterize sediment trajectories and persistence and quantify impacts to SG. This knowledge will assist resource managers and stakeholders to develop guidelines that protect SG resources and sustain oyster harvest.

We'll study an operating COM to quantify the sediment generating activity, then devise a small-scale simulation to apply within SG beds. Sediment plumes will be generated by walking in an unvegetated patch for specified time and distance. Plumes will be characterized relative to sediment type, wind/current velocity, time and distance to quantify sedimentation outcomes. The disturbance protocol will be repeated 1-2X/wk to mimic a COM while monitoring sediment transport and SG condition/quality. The primary outcome will be established relationships between disturbance activity, sediment transport/plume characteristics, and SG status, as a test of the 200 ft buffer efficacy. Success will be adoption of this knowledge for science-based siting decisions.

Objectives

Obj. 1: Quantify sediment generating activity at existing COM operation to gauge plume characteristics; Then devise an applicable simulation model.

Obj. 2: Perform controlled experiments to characterize sedimentation profiles (sediment loads, light attenuation with distance/time from disturbance event) for various environmental factors (sediment type, current velocity), and quantify relationships between activity, physical conditions and sediment plume trajectory, persistence and spatiotemporal variation.

Obj. 3: Utilize above results to develop standard disturbance protocols and apply them repeatedly as in an ongoing COM operation; Monitor abiotic indicators for cumulative sedimentation effects, and biotic indicators to show how sedimentation levels impact SG condition and quality.

Obj. 4: Use innovative methods to: A) Monitor sediment plume characteristics by video and time-lapse photography from Unmanned Aerial Systems (UAS); B) Establish SG “condition” spatiotemporally by UAS imagery of a time series of SG areal distribution, along with SG biomass and morphological indicators; and C) Determine SG quality via its contributions to its ecosystem: algal epiphytes and benthic invertebrates are food for higher consumers. Epiphyte communities will be assessed by image analysis of color-scans; Benthic invertebrate sampling will assess their abundance and community diversity.

Obj. 5: Analyze project data to establish overarching relationships between sediment generation and SG status to assess efficacy of buffer zone size on SG protection. Interpret data with input from agency staff, researchers and stakeholders, and widely disseminate findings.

This project will provide critical data on the quantitative relationships of COM-SG interactions. Beneficial economic impacts derive from productive COM that promotes SG sustenance of other marine resources. We received valuable input/suggestions from TPWD staff (Mr. Olsen, Dr. Clarkson, Dr. Campbell).

Tasks

Task 1: Pre-Experiment Preparation

TAMUCC will (1) quantify parameters to develop an appropriate sedimentation simulation that mimics observed COM operations, (2) identify suitable study sites, and (3) develop a preliminary Quality Assurance Project Plan (QAPP) for experimental and analytical procedures. TAMUCC will develop the sediment simulation based on observations of existing pilot studies and COM operations with simultaneous UAS (small unmanned aircraft systems) and ground-level video recording. The recordings will quantify any sediment disturbance by type (wading impacts, equip-ment, boat), duration, frequency, and impact area. SG maps and Texas Parks and Wildlife Department (TPWD) staff will be used to identify potential sites representing different SG sediment types (sandy or muddy). Preliminary UAS surveys, physical site measures, and grab samples will be used to evaluate and compare candidate sites.

Task 2: Study Design and Experimental Plots

TAMUCC will select two SG study sites representative of muddy and sandy sites. Each site will have three plots: no disturbance, low intensity disturbance, and high intensity disturbance.

TAMUCC will finalize the QAPP and the experimental design using input from TPWD and mariculture/SG experts. TAMUCC will mark the study sites and measure the pre-disturbance parameters in all plots.

Task 3: Experimental Data Collection

TAMUCC will complete the disturbance experiments over approximately three months (mid-May to mid-August). During the disturbance experiments, TAMUCC will use UAS equipped with high-resolution RGB and near-infrared cameras to collect the sediment plume data and areal SG coverage. TAMUCC will also complete synchronized monitoring of plume characteristics (wind/current speed, light availability logging with Hobo light meters, sediment traps, turbidity). TAMUCC will sample SG shoot density, morphology (leaf length, leaves/shoot), and biomass within each plot every two weeks. TAMUCC will take sediment cores for root/shoot indicator ratios at the project start and end. TAMUCC will quantify the SG epiphyte community biweekly using biomass and leaf coverage estimated with ENVI software. The epiphyte community composition will be characterized to the extent practicable using spectral libraries for any distinguishable classes of algae, and quantifying coverage by class. TAMUCC will sample and identify benthic macroinvertebrates to the lowest practicable taxon.

Task 4: Data Analysis, Reporting, and Dissemination

TAMUCC will analyze the UAS imagery of sediment plumes and SG, as well as abiotic/biotic measures, relative to sediment type, disturbance intensity, wind/current velocity, and distance/time from the disturbance event (physical conditions). TAMUCC will seek data interpretation input from the Texas Seagrass Monitoring Working Group, TPWD and mariculture/SG experts, and other COM stakeholders. TAMUCC will complete the data analysis and share significant findings in a final report. TAMUCC will share the final report publicly via a website developed by TAMUCC, Cyverse for seagrass imaging data, peer-reviewed publications, and conference presentations.

Task 5: Project Monitoring & Reporting

TAMUCC will prepare and submit all reports, deliverables, and requests for reimbursement as required in the contract, to CMPReceipts@GLO.TEXAS.GOV. Quarterly progress reports and requests for reimbursement are due to CMPReceipts@GLO.TEXAS.GOV on the 10th day of every quarter of the year starting with January 10, 2022.

TAMUCC will provide a final report describing work completed under each task.

METHODS

Experimental Design – Seagrass Plot Arrangement

The experimental design facilitated comparisons between each of 3 different levels of sediment disturbance at sites with extreme differences in ambient sediment conditions (ie muddy vs sandy sediments). Thus treatment levels can be compared within sites (ie between tracks), and equivalent treatment levels between sites. Moreover, the characteristics of the sediment plumes, and any impacts on seagrasses, were measured and analyzed as a function of distance from the sediment plume source to test the efficacy of the existing 200 ft buffer zone required between the COM operations and seagrasses.

Each site was subdivided into 3 “tracks”, approximately 100 ft (30 m) wide (perpendicular to predominant current direction) by 200 ft (61 m) in length (parallel to current direction). Each track had a different degree of sediment plume disturbance: Control (Track 1; no disturbance), a relatively low level of disturbance (Track 2), and a relatively high level of disturbance (Track 3).

The disturbance path was located at the baseline of each track. The pathlength was at 50 ft (15 m), typically centered in each 30 m wide track. Measuring stations were centered in each Track at varying distances, typically 50, 100 and 200 ft (15, 30 and 61 m) to provide a reproducible location for measurements and sample collections. The Site and Track setup is illustrated in Figure 1.

A variety of physical parameters were measured to characterize the sediment plumes, and both biotic and abiotic sampling took place as a function of distance from the sediment plume source.

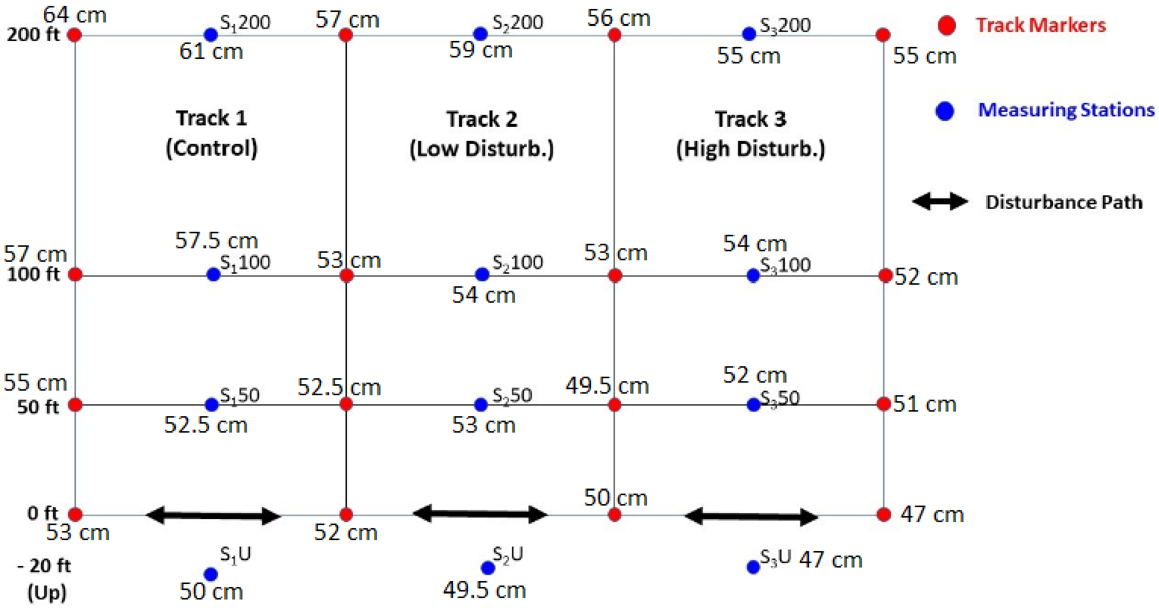


Figure 1: Seagrass Study Site Layout. Note measuring stations and disturbance path at the baseline. Measuring stations labeled with name for samples taken at each location. “S” represents samples taken at San Jose area; Turtle Bayou samples would be labeled with a “T”. Depth marked at each pole in centimeters

The sediment disturbance treatments attempted to replicate plumes of similar intensity and size based on our measures at COM operations. These plumes were generated by walking along the up-current edge of the plot, in a standardized fashion, to kick up sediment plumes. These would simulate a shallow, wading-based maintenance operation at a COM site. Different levels of disturbance intensity and duration were determined after observations were completed of sediment plume generation at COM sites. Preliminary studies consisted of, 5-, 15- and 30-min disturbance periods, where the disturbance consists of a person walking back and forth along a 50 ft (15 m) long path centered at the up-current edge of each track. But the intention is for disturbance levels to bracket those observed at COM sites. The calibration of sediment disturbance mimicry was based on trial-and-error disturbances at seagrass plots with simultaneous measurement of sediment plume characteristics (see below). These measures included light attenuation, Total Suspended Solids (TSS), sediment dry weight settled in sediment traps, and image-analysis-based information processed from UAV aerial imagery.

The disturbance path was located at the baseline of each track. The path length was 50 ft (15 m), typically centered in each 30 m wide track. However, wind-driven currents shifted orientation to different degrees, so the disturbance path was adjusted, to allow suspended sediment plumes to stay within the boundaries of the tracks and reach the measuring stations positioned at 50, 100 and 200 ft (15, 30 and 61 m) down-current.

Study Sites

Commercial and Research COM

Four oyster mariculture sites were visited on multiple occasions each for observation of mariculture procedures and measurement of sediment plume parameters. We had planned to visit the other two commercial sites that were permitted at the time (at Tres Palacios and Galveston Bays) but were unable to schedule a visit because of our time constraints and logistics. In all cases, the responsible operators and permit holders were extremely cooperative and willing to provide us with assistance. We had contacted and discussed visiting Mr. Aparicio's Tres Palacios farm in Summer 2022 but we decided to delay the visit since they were just beginning operations and we were having difficulties in other aspects of this project. We later discussed a visit there in March 2023 but our restricted availability did not mesh and the project was nearing the completion date. The sites utilized in this study were 3 shallow water (wadable from shore; 3-4 ft deep) research sites that used suspended cages or the Adjustable Long Line system,, and 1 operational commercial site that was considered deep water (8 - 10 ft depth only accessible by boat). These sites are described below.

Shallow Water Research COM Sites

Corpus Christi Bay – Hollenbeck Research COM Site

[1]

A small oyster mariculture research operation was on TAMUCC property off Ward Island in Corpus Christi Bay. This facility was operated by TAMU-CC and Texas A&M AgriLife-associated faculty Dr. Chris Hollenbeck. This operation was only 1 line of 8-10 cages, and another smaller line of 3-4 cages. The cages were accessed by wading from shore (3-4 ft depth). The substrate at this location was primarily firm bottom sand, with patches of soft mud or oyster bio deposits directly underneath the cages. Seagrass was not present here or at any location under direct hydrological influence from the mariculture research operation. Current at this site was variable due to eddies created by the Jetty's presence on the eastern side of the cages, and prevailing westerly current in deeper water, alternating with winds and current out the North during cold fronts. UAS imagery was not possible at this location due to its location in a restricted airspace.

Copano Bay – Harte Research Institute (HRI) Research COM Site

This shallow water (3-4 ft depth) research mariculture facility operated by HRI was located along the southern shore of Copano Bay. It had a firm sandy bottom with soft mud pockets directly adjacent to or underneath the mariculture equipment. According to a previous study done here (Rubino 2021), there was some sparse *Halodule* wrightii seagrass around the operation, primarily concentrated in shallow guts created by the high wave energy. The water was never sufficiently clear for us to observe any seagrass presence, and we did not encounter it when anchoring our logger poles and sediment traps. This site used three adjustable long line systems with suspended baskets organized in parallel with each other. No mariculture operations were active at this site as of July 2022 having been recently closed, so we simulated any potential operational impacts from wading (described below).

Tres Palacios Bay – HRI Research COM Site

Another HRI shallow water research COM facility was located along the northern shore of Tres Palacios Bay and was the most active of the 3 shallow sites, although this site too was winding down operations which ceased after August 2022. It once had 3 cagelines with each capable of holding about 30 floating cages. At the time of our visits, one line of about 30 cages remained. The substrate matched up similarly with the other two shallow sites, with a firm sandy bottom, and pockets of soft muddy sediment underneath the mariculture equipment. Depth typically ranged from 3 – 5 ft, and operators accessed the cages by wading out with a floating platform to transport gear and oyster sacks. For our work, we utilized both the platform and kayaks, with the latter being faster, easier and less disruptive to the bottom when shuttling our measuring equipment to different stations around the facility.

Deep Water Commercial COM Sites

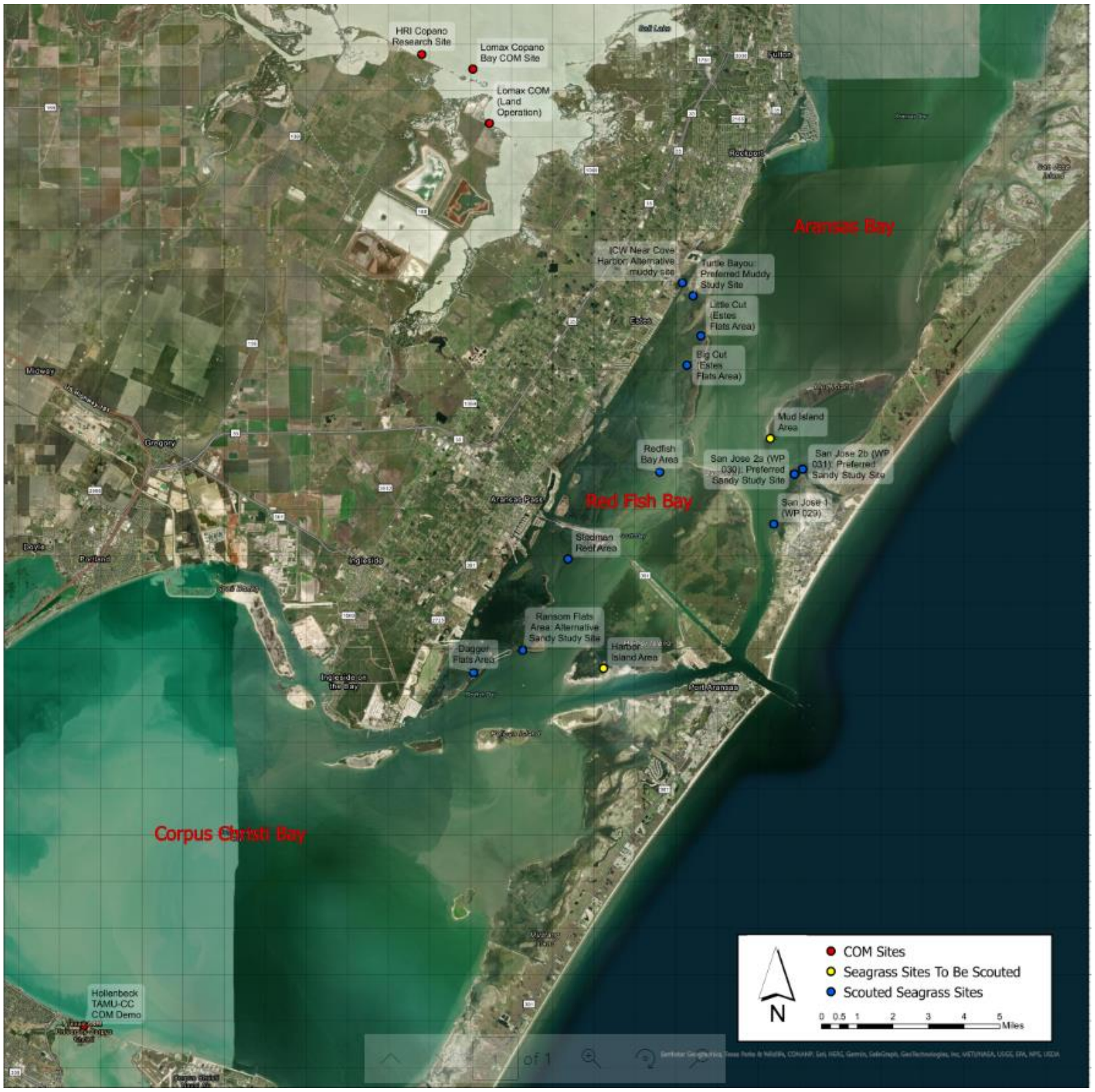
Only one deep water commercial scale COM facility was able to be surveyed for this project due to issues with the logistics of scheduling sampling.

Copano Bay – TX Oyster Ranch (Lomax)

The first permitted COM in TX, the Texas Oyster Ranch is located along the southeastern edge of Copano Bay near Swan Lake. This mid-scale commercial site is in relatively deep water (8-10 ft deep) and only accessible by boat. The bottom is predominantly soft mud near the cages, but of unknown (to us) character outside the immediate farm area. However, towards the shoreline to the south, the water becomes shallower with a sandy bottom. There were 7 rows of up to about 36 cages per row for commercial production, plus an eighth row with some research cages (Dr. Hollenbeck) at the far western end (not visible in picture above). TX oyster Ranch employees provided boat transport for researchers to deploy instruments, measure water and sediment plume parameters, launch UAS flights, and observe COM procedures.

Seagrass Beds

Scouting



Study sites for the seagrass part of the experiment were scouted during December 2021- April 2022. 12 potential sites within Red Fish and Aransas Bays were visited and assessed for potential suitability for a seagrass study site location following the guidelines shown on the accompanying map (above; marked by blue dots). Red dots on the map mark locations of COM study sites at various locations in Corpus Christi Bay and Copano Bay. The Tres Palacios Bay research and commercial COM sites are not shown in this map.

The map below shows the final selected study sites for observing COM operations and the two seagrass sites selected for use in the disturbance experiments. Turtle Bayou, near Rockport, and San Jose along the western edge of San Jose Island, southeast of Mud Island. The Tres Palacios Bay research and commercial COM sites are not shown in this map.

GPS coordinates for each site are given in the table below.

	X	Y
Turtle Bayou (muddy study site)	-97.076433	27.976566
San Jose (sandy study site)	-97.03087832	27.91386258
Lomax Deep Water COM Site	-97.165752	28.057717
HRI Copano Shallow Water COM Site	-97.186506	28.062988
Hollenbeck Shallow Water COM	-97.3236121	27.7148957
HRI Palacios Shallow Water COM	-96.23948742	28.69214712

Seagrass Study Site Selection

Two seagrass study sites were selected based on the following criteria:

- Primarily *Halodule wrightii* seagrass beds
- One site with more sand-based sediment, and another site with more silt and clay based sediments
- Consistent seagrass coverage over an area of approximately 100 m x 100 m
- Consistent depth across chosen area
- Sites oriented approximately square with prevailing SE winds (to produce reliable wind-driven currents)
- Sites bounded on the upwind/upcurrent side by unvegetated sediments or low-cover seagrass facilitated a sediment disturbance protocol (see below)

Candidate sites were identified through Google Earth imagery and examined by boat, depth measurement, and by aerial UAV imagery. Georeferenced imagery was used to verify the layout of experimental "tracks" (see below).

Selected Seagrass Study Sites

After repeated scouting of candidate sites for the seagrass study, two preferred locations were chosen in accordance with the criteria listed above. One being a shallow seagrass bed of predominantly *Halodule wrightii* with a firm sandy substrate located on the western side of San Jose Island southeast of Mud Island. The second site was a shallow bed of *Halodule wrightii* (and some *Ruppia maritima*) with a softer, muddy-based substrate located in Turtle Bayou near Rockport, TX.

San Jose Island

The first Seagrass study site that was selected was located on the western side of San Jose Island Southeast from the Southern tip of Mud Island, near North Pass. Depth at this location varied by up to 14 cm across the entire plot (mean depth 40 cm). A study site map showing depths across the site is presented below. This site served as the seagrass study site with a sandy substrate. *Halodule wrightii* was the predominant species, spread in a uniform pattern and density across the entire experimental plot. The experimental plot was rotated to the Northeast with the baseline following the compass heading 30 degrees ~NE. This was done to match prevailing wind patterns in the area coming out of the Southeast. Weather forecasts and conditions for this site were obtained from using the Port Aransas (Inside) weather station on Willy weather.

Turtle Bayou

The second, muddier seagrass study site was in Turtle Bayou on the northwest side of a small channel at the north end of Estes Flats of Redfish Bay near Cove Harbor marina. The substrate was a soft, mud-based sediment. Average depth here was 40 cm ± 10 cm. The current at this location appeared to follow a small boat channel into Aransas Bay to the east. *Halodule wrightii* was the dominant species of seagrass in this bed, but *Ruppia maritima* was also present across the experimental plot. The baseline for the experimental plot followed a compass heading 60 degrees ENE to account for prevailing southeastern winds. Weather conditions and forecasts for this site were obtained from the Rockport weather station on Willy Weather.

Field Measurements and Collections

Site and Environmental Conditions

During all sampling trips during the project, several environmental conditions were measured and recorded to keep as a log in efforts to correlate intensity and direction of sediment plume travel with conditions at the time of experiment. These conditions included the frequent measurement of both wind speed in both meters/second and miles/hour using weather station data and data collected using an anemometer during field sampling procedures. Wind direction was recorded. General weather and water conditions were noted routinely, as well as cloud cover, relative water depth, wave activity, and general turbidity. The tide activity was also obtained from using weather forecast websites such as Willy weather. Salinity was recorded at each visit as well, ranging from 30-40 ppt throughout the duration of the project. Current was measured at each sampling visit by using either SonTek Flow Tracker 2 Acoustic Doppler Velocimeter, or Hach FH950 Portable Velocity meter. Wind and current conditions were measured regularly in efforts to document any variation or changes in current or wind patterns that could affect sediment plume travel.

UAS Aerial Imagery

High-resolution images of the selected seagrass sites were collected using unmanned aircraft systems (UAS) that were equipped with RGB or infrared cameras. Following FAA regulations, the UAS flights were conducted using a DJI Mavic Pro 2 and a DJI spark at no more than 120 m (400 ft) above the water surface and consist of multiple types of operations.

During site scouting, the flights were manually controlled by a certified FAA UAS pilot, with potential assistance from a visual observer. The pilot made real-time decisions on when and where the images will be taken, based on communication with the ground team.

After the sites were selected, the flights for disturbance monitoring on a regular basis included two different modes. In the static mode: the UAS were deployed at a high position so that the entire 300 x 200-ft site (3 Tracks) was covered in one scene, in order to save battery life and extend flight time. In the tracking mode, the UAS flew at a lower altitude and followed the movement of the sediment plume from the disturbance location through the seagrass bed, observed the motion, expansion, and persistence. In both the static and tracking modes, centimeter-level images and/or videos were produced. Ground control points were deployed at each site as visual indicators in aerial imagery.

After every flight, UAS data was collected and immediately transferred to a field laptop. After each trip, the data was moved to a workstation for image analysis and uploaded to an online database for sharing with team members.

Physical Parameter Measurements to Characterize Sediment Plumes and Site Conditions

Sediment plumes and general site conditions were documented and quantified by measuring physical parameters: Water temperature, depth, salinity, wind direction and speed, water current direction and speed, tide readings from the nearest gauges at Port Aransas (Inside), Rockport, and Ingleside On The Bay. Light attenuation was measured by light loggers deployed during sampling periods.

Abiotic sampling measured Total Suspended Solids, sediment dry weight settled in sediment traps, and sediment grain size distribution and organic content.

These measures informed image-based analyses of pictures and videos recorded by UAV. Image-based metrics will be developed.

Physical and Environmental Parameters

During sampling, field environmental conditions including water depth, salinity, temperature, weather, water, and wind observations were recorded. Depths were taken with a calibrated PVC pipe. Temperature was taken at canopy height with a calibrated thermometer, or else recorded by the Hobo Light Loggers (MX 2202, Onset Computer Corp.). Water samples were collected at canopy height into 50 ml Falcon tubes without headspace. These samples were taken to the laboratory where salinity was measured with a refractometer (VEE GEE, STX-3) at a consistent laboratory temperature (~23C) to avoid improper temperature compensation when field water temperatures exceeded 30° C. Before testing, samples were stored at 4° C. These samples were equilibrated to laboratory temperature for at least 4 hours. The refractometer was calibrated with equivalent temperature deionized water.

Weather observations (including historical) and wind and water conditions (Tides) were obtained from web-based sources such as Wunderground (https://www.wunderground.com/history) and Willy Weather (https://tides.willyweather.com/tx/nueces-county/port-aransas-inside.html).

When documenting sediment plumes, additional parameters were critical for data interpretation. These included wind speed and direction, water current speed and direction, Total Suspended Solids (TSS), and settled sediments collected in sediment traps. Wind speed ranges at various times were measured with an anemometer. Limited water current measurements were obtained using a SonTek Flow Tracker 2 (Acoustic Doppler Velocimeter) that was rented for July 2022. Manufacturer's instructions were followed (https://www.geotechenv.com/Manuals/SonTek_Manuals/sontek_flowtracker2_manual.pdf). At other times, current measures were acquired using a Hach FH950 Portable Velocity meter.

In some cases, these measurements were taken at varying distances from the source of the sediment plume disturbance.

Sediment Composition Analysis

The sediment composition (grain size distribution, organic content) is an important factor in seagrass productivity. These factors were characterized at each track of both study sites pre- and post-disturbance.

Three intact sediment cores (3 cm diameter, 10 cm deep) were collected from near each measuring station in each Track, composited by placing into a plastic bag and stored on ice.

Upon return to the laboratory, cores were stored at 4° C until processing (not more than 1 week). Alternatively, cores can be stored frozen at –20° C.

Sediment Plume Parameters

The sediment plumes were characterized by the amount of suspended materials they contained, as well as their relative proportions of organic vs inorganic constituents.

Water samples for Total Suspended Solids (TSS) were collected into clean 0.5 L or 1 L plastic bottles which were first pre-rinsed 3 times w/ambient site water. Sample collection was made by immersing the bottle to slightly above seagrass canopy depth and opening the lid to fill the bottle without headspace. Samples were placed on ice and kept dark. Upon return to the lab, samples were transferred to storage at 4° C. Samples were processed within 72 hrs.

Sediment traps allowed settleable solids to be collected, dried and weighed. These devices were similar to those used previously (Rubino 2021) and comprised a funnel (10.2 cm diameter) glued into a 3” diameter PVC fitting that contained a collection jar. The traps were deployed on the sediment bottom and fixed in the upright position by attachment to a piece of 3/8” re-bar. The mouth of the funnel is approximately 13 cm above the sediment. Traps were placed at each measuring station 30 min prior to sediment disturbance and collected at least 30 min following the sediment disturbance. Caps were fitted to the collection jar and samples stored dark and on ice for transport to the laboratory. Upon return to the lab, samples were stored at 4° C and processed within 72 hrs.

Sediment Trap construction and collecting jar that is inserted inside. The area of the collection funnel is 0.011 m^2. This is anchored upright on the bottom by threading a piece of rebar through a hole in the funnel tab.

Light Logger Pole showing the “Air” logger at the top, and a pair of loggers separated in depth by 13 cm, and the lowermost logger sits 10 cm off pf the bottom. This is placed over a piece of rebar inserted vertically in the sediment to hold upright. A long tent peg is inserted into sediment through a hole in the lowermost horizontal PVC to prevent turning in the current.

Floating Measurement Station for use at deep water COM sites. Light Logger Poles and Sediment Traps are zip-tied into proper orientation on a concrete reinforcing wire mesh. A 2 pound weight at bottom of mesh keeps the mesh vertical in strong current. Floats attach to the upper corners of the wire mesh and are adjusted to position top of wire mesh at 2 ft beneath the surface. Half-concrete block anchors (not shown) also attach where the floats do. A longer pole is added to mount an "Air" logger above the water surface.

A floating measurement station deployed at the Lomax deep water COM Site.

Light Attenuation

A consequence of sediment plumes is the attenuation of light penetrating the water column and available for seagrasses. Two methods were used to quantify and study both ambient light conditions and attenuation of light by sediment plumes. LiCor spherical sensors are typically used to measure photosynthetically active radiation (PAR; µmoles photons/m^2-Sec) levels available at various depths in the water column and to calculate a light attenuation factor (k) and the percent surface irradiance available at the seagrass canopy (Dunton, 2018). The setup uses two spherical sensors at different fixed depths, mounted on a pole and cabled to a data logger which was not waterproof. The difference in light readings with depth provided a measure of the light attenuated by suspended sediments and phytoplankton. However, these were point in time measurements and also cumbersome to deploy unless on a boat or platform. Moreover, the shallow water depths at the *Halodule* seagrass beds in this study limited use of this method. It was sometimes used in water > 3ft deep when a light logger pole was also deployed.

An alternative method used Hobo MX2202 light loggers (Onset Computer Corp) which measured and recorded light (and temperature) over time intervals up to days. This quantified light levels over time before, during and after a sediment plume passed by. Thus, we could correlate light attenuation with passage of a sediment plume and calculate a “light budget” at each deployment location. A limitation of this approach was that the Hobo light loggers use different technology, and measure light at green wavelengths which are not used efficiently by seagrasses, and the units (Lux) are not directly convertible to the units of PAR (µmoles photons/m^2-Sec). Nonetheless, suspended sediments were expected to be relatively neutral in the light wavelengths attenuated (ie block most wavelengths), so the wavelength is less critical because large particle blockage of green light likely also blocked PAR.

Two Hobo light loggers were attached to horizontal PVC arms affixed to a vertical PVC pole at two different depths. For shallow water applications, the pole was positioned so that the bottom logger was 4 cm off the bottom (in the seagrass canopy, which was locally clipped to prevent interference). The upper logger was fixed at approximately 13 cm above the position of the lower one. The difference in light levels recorded by the two loggers corresponds to the light attenuation by a 13 cm column of ambient water. The light logger poles were deployed at each measuring station by guiding the PVC pole over a vertical piece of re-bar so that the logger arms were positioned due south of the support pole. A large 8” metal spike (tent peg) was used as a secondary anchor point on the lower arm to prevent currents from rotating the logger pole and altering their relation to the sun’s position. The light logger poles were deployed at least 30 min before a sediment disturbance and removed at least 30 min after a sediment disturbance.

Where the water was sufficiently deep, we performed several simultaneous side-by-side deployments of the LiCor and Hobo systems to establish a correlation. Plots of available light over time indicated changing turbidity by passing sediment plumes, which were expressed as the difference between the upper and lower loggers divided by the upper logger reading. This allowed calculation of the Percent Relative Difference to represent light attenuation by the sediment plume.

Biotic Sampling to Monitor Seagrass Bed Conditions

Biotic parameters indicative of seagrass system functioning were sampled at the chosen sites prior to initiation of regular standardized disturbance regimes, and then measured periodically (approximately once every 3 weeks) throughout implementation of the treatment regime, and at the endpoint. Destructive sampling that digs up seagrass rhizomes were minimized as much as possible.

Seagrass sampling comprises harvesting 15 haphazardly selected *Halodule wrightii* shoots for targeted seagrass indicators that included shoot morphometrics (leaf length, leaves per shoot), average biomass per shoot, average epiphyte biomass per shoot, and imaging-based leaf coverage by epiphytes. Such samples were taken at varying distances (typically 50, 100 and 200 ft, or 15 m, 30 m, and 61 m) from the sediment plume disturbance, for each of the treatment tracks. These parameters were typically measured every three weeks during the disturbance sampling.

Seagrass shoot densities were compared during the disturbance phase by “Ring Sampling”. This method entailed collection of all seagrass shoots located within haphazardly placed (3 per measuring station) 2” inner diameter PVC rings (0.00203 m^2 area). This harvest method left the seagrass rhizomes and sediments intact.

Seagrass core sampling to 10 cm depth took place twice using a 10.5 cm diameter corer: pre-disturbance and again at the endpoint. Triplicate seagrass core samples were lab-processed to record below-ground to above-ground seagrass biomass ratios. These same core samples were also used to assess the macroinvertebrate community.

Macroinvertebrates associated with seagrasses served as indicators of seagrass system functioning. These were monitored twice as for the seagrass cores. To minimize seagrass bed disturbance, the same cores were used for both seagrass and macroinvertebrates.

Core Sampling for seagrass belowground and aboveground biomass.

Seagrass and Invertebrate Bioindicators

Seagrass and invertebrate bioindicators were sampled during the sediment disturbance part of the experiment, as well as pre- and post-disturbance. Core samples enabled simultaneous analysis of seagrass and fauna, but its destructive nature means it was limited to just pre- and post-disturbance analyses. Instead, regular sampling during the disturbance period were conducted by seagrass shoot-harvesting and faunal sweep-net methods.

Seagrass shoot sampling during the disturbance phase comprised of harvesting 15 haphazardly selected *Halodule* shoots. These were analyzed for targeted seagrass indicators such as shoot morphometrics (leaf length, leaves per shoot), average biomass per shoot, average epiphyte biomass per shoot, and imaging-based leaf coverage by epiphytes.

Seagrass shoot samples were collected by pinching the vertical shoot below the leaf attachment point and placing them into plastic bottles and transported *on top of* ice and stored at 4° C - 10° C in the laboratory. Initial sample processing in the laboratory were completed within 72 hours of harvest.

In addition, seagrass shoot densities were compared during the disturbance phase by "Ring Sampling". This method entails collection of all seagrass shoots located within haphazardly placed (3 per measuring station) 3" inner diameter PVC rings (0.0082 m^2 area). This method left the seagrass rhizomes and sediments intact.

Core sampling for both seagrass and invertebrate indicators were collected before the disturbance protocol is initiated and again after the disturbance protocol has finished. The cores (10.5 cm diameter; 10 cm deep) were used to compare seagrass below-ground to above-ground biomass ratios and faunal diversity and abundance.

Core samples were taken near each measurement station within each Track, at positions that are relatively undisturbed by other sampling activities. The corer was placed over the seagrass bed haphazardly and twisted to cut through seagrass shoots and rhizomes while pressing downward to the correct depth. Care was taken to collect only intact core samples. Core contents (seagrass, fauna, and sediment) were brought to the water's surface and deposited into a sieve with 1mm mesh. Contents were sieved in the field and seagrass plants removed for assessment. Seagrass shoots and rhizome/roots were collected into plastic bags and stored on ice in a cooler. After sediment is largely sieved away, invertebrate fauna were gently washed into a plastic collecting jar or picked by forceps. A vital stain and formalin were added to preserve the fauna. Jars were returned to the lab for taxonomic identification to the lowest practicable taxon.

Seagrass samples collected in the field were stored at 4° C in the laboratory.

Laboratory Methods

Processing UAS Data

The UAS data was processed using the Motion (SfM) technology in ArcGIS Pro and Pix4D Mapper. The analysis included several steps: keypoint extraction, keypoint matching, camera optimization, geolocation, point cloud, 3D textured mesh, and image mosaicking. The accuracy of spatial referencing was enhanced by incorporating the measurements of ground control points. The results included a set of site-scale orthomosaic images at a spatial resolution of up to 10 cm. These images were used for the evaluation of seagrass coverage and the characterization of sediment plumes.

The UAS data and derived geospatial products were also uploaded to an online platform for the dissemination of the project results. This platform were developed using the ArcGIS AppBuilder technology to support the management of both vector and raster datasets.

Metadata of the geospatial products were prepared according to ISO 19115 to ensure both the readability for the general public and the compatibility with international standards. All geospatial data was assessed for quality, completeness, and relevance using standards identified from literature review. The assessment emphasized geometric accuracy, consistency of spatial resolution, and user accessibility.

Measurement of Seagrass Indicators

Seagrass samples collected in plastic bottles or bags are placed on top of ice for transport and stored at 4° C - 10° C in the laboratory. Initial processing took place as soon as possible, within 72 hrs of collection.

Seagrass shoot samples consisted of 15 whole shoots haphazardly selected from sampling locations within each of the two study sites: sandy and muddy. These were processed for a standard set of seagrass bioindicators:

- Average number of leaves per shoot
- Average leaf length
- Seagrass biomass
- Epiphyte dry weight biomass
- Epiphyte ash weight
- Epiphyte community fluorescence characteristics
- Leaf coverage by epiphytes (imaging based)

Seagrass “ring samples” (all shoots in the 0.002 m^2 PVC ring) for shoot density were counted and expressed per unit area.

Seagrass recovered from core sampling were used to compare below-ground to above-ground biomass ratios. Field processed seagrass samples in bags were transferred to DI water, dis-entangled and picked free of shells and other debris. Plants, and parts thereof, were separated into above-ground (vertical shoots and leaves) and below-ground (rhizomes, roots) portions. The above- and below-ground portions were dried to constant weight at 60° C to calculate the biomass ratio.

Seagrass Imaging and Morphometric Measurements

Seagrass samples collected in plastic bottles were placed on top of ice for transport and stored at 4° C - 10° C in the laboratory. Initial processing takes place as soon as possible, within 72 hrs of collection.

Seagrass samples were gently soaked for 2-3 minutes in deionized water to remove salt and loose, non-specifically associated material (e.g. mud, sand). For each single whole shoot, the green portions of leaves were excised from the junction with the leaf sheath, measured for morphometrics (leaf length, number of leaves per shoot), and then arranged on a fluid mount scanning tray for the Epson Perfection V-750 Pro color flatbed scanner (Epson, Carson, CA). Leaf images were obtained using 24-bit color scanning at 1200 dpi resolution and then saved as .tiff files. Scans were performed twice, once each with a black background, and again with a white reflective background.

After scanning the leaves, epiphytes were removed from seagrass blades by gently scraping with a microscope slide into a small volume (<50 mL) of deionized water. The epiphytes are ground in a mortar and pestle, quantitatively transferred to measure volume, and then 1 mL aliquots (3) are taken for subsequent assays which included fluorescence and pigment analyses. One aliquot is stored at 4° C for fluorescence measurements within 48 hrs. Two aliquots are stored at -20° C for other optional measurements such as pigment analyses or DNA extractions. The epiphyte-free leaves and remainder of removed epiphytes were then dried to constant weight at 60° C for biomass measurements.

Biomass and Ash Weight Measurements of Seagrasses and Epiphytes

After scanning the leaves, epiphytes were removed from seagrass blades by gently scraping with a microscope slide. The epiphyte-free leaves and remainder of removed epiphytes were then transferred to pre-weighed beakers and aluminum dishes, respectively, and dried to constant weight at 60° C for biomass measurements. Three consistent biomass measurements (exhibiting only small variations due to ambient humidity) were required before proceeding to acquisition of the ash weight of the epiphytes.

Dried and biomassed dishes of epiphytes were transferred into a muffle furnace and ashed at 550° C for 2 hrs (at ashing temperature). Careful re-weighing gave the inorganic weight, allowing calculation of the relative proportions of the epiphyte dry weight that are organic and inorganic. It was hypothesized that epiphyte fractions from high suspended sediment environments would exhibit greater proportions of inorganic content (sand).

Fluorescence Characterization of Epiphyte Communities

After seagrass scanning, removed epiphytes were homogenized in a mortar and pestle, volume measured and aliquots stored at 4°C for fluorescence characterization within 48 hrs. Portions (200 µL) of these epiphyte samples were transferred, in triplicate, into a black-bottom 96 well plate using a cut micropipette tip to accommodate suspended pieces of macroalgae. Fluorescence measurements were acquired with multiple excitation and emission wavelengths to detect chlorophylls and phycoerythrins using a Thermofisher Varioskan LUX multi-mode plate reader with Skan-It Software v4.1. A standard measurement template was used to specify top-read of fluorescence emission for 100 ms. The multiple excitation and emission wavelengths were described in the Table below. Excitation bandwidths are 15 nm.

Excitation 1 (nm)	Excitation 2 (nm)	Excitation 3 (nm)	Excitation 4 (nm)	Emission (nm)
415	500	530	565	680
415	500	530		576
			565	620
			565	655

Fluorescence emission results were loaded into an analysis template that calculates the ratio of phycoerythrin fluorescence to chlorophyll fluorescence (530 Ex – 576 Em) / (415 Ex – 680 Em); called “R/G Ratio”). This value was used as a general indicator of epiphyte community composition. It detected large shifts between dominance of the community by red or green algae. Chronic changes in light quantity or quality, a potential result of suspended sediments, may shift this ratio.

Processing Sediment Composition Samples

Three replicate sediment cores (3 cm diameter, 10 cm deep) were collected from each track and composited or stored at 4° C or –20 ° C until processing (not more than 1 week). Sediments were processed using a dry-sieve method (Folk, 1980). In this case, composited samples were thoroughly mixed and dried at 60° C. The dried sediments were transferred to a pre weighed beaker and then reweighed to calculate total sediment dry weight. Dried sediments were then transferred to a series of stacked, nested sieves of sizes 1000 µm, 500 µm, 250 µm, 125 µm, and 63 µm over a collection pan. The stack was secured onto a RO-TAP Sieve Shaker and sieved for 15 min. Sediment fractions were recovered by inverting sieves onto a large sheet of clean paper and gently tapping diagonally to the mesh. Collected sediments were transferred into pre-weighed aluminum dishes. Remaining sediments in the sieve and on the paper were quantitatively recovered by inverting the sieve and pounding the rim sharply, but evenly, onto the paper. A brush was used to remove any remaining particles. All recovered fractions were weighed, and then ashed twice for 2 hours at 550° C and re-weighed. The weight change represents the organic proportion, and the ash weight represents the inorganic proportion. The proportion of total sediment dry weight *recovered* that is comprised by each size class, as well as proportions of organic and inorganic constituents, were compared between sites and tracks.

Sediment Traps

Settled sediment samples recovered from Sediment Traps had been collected into 130 mL jars. These were stored at 4° C and then processed within 72 hours after collection.

Measurement and analysis of sediment trap samples began with carefully decanting off cleared water from undisturbed jars into a 200 mL vacuum filtration funnel containing a pre-weighed 1.2 µm Glass Fiber Filter (VWR 696) and filtering. The majority of the sediment remaining in the jar was then resuspended in the small remaining volume of liquid and DI water was added to refill the jar to dilute the salt content of the native water. This resuspension of sediment in DI water was allowed to resettle and then decanted into the filtration funnel a second time leaving the sediment in a small volume of water at the bottom of the jar. This small volume was then added slowly to the filtration apparatus via pipette to quantitatively transfer all remaining liquid and sediment, rinsing with DI water as needed. After filtration, filters with all collected solids were transferred to pre-weighed aluminum dishes and dried to constant weight at 60° C. These were weighed, and then typically further ashed for 2 hrs at 550° C for a final weight to determine the organic and inorganic proportions of the settled sediments.

Total Suspended Solids (TSS)

Water samples for TSS (0.5 L or 1 L) stored at 4° C will be processed within 72 hrs.

Measurement of the TSS began with recording the sample volume and allowing heavy solids to settle for at least 4 hrs. Quantitative sediment collection started by decanting the cleared water into a vacuum filtration funnel and filtering through pre-weighed 1.2 µm Glass Fiber Filters (VWR 696). Settled solids were suspended in the small volume of sample liquid remaining and quantitatively transferred into the filtration apparatus. DI water was used to rinse all vessels holding the sample. Filters with all collected solids were transferred to pre-weighed aluminum dishes and dried to constant weight at 60° C. These were weighed, and can be further ashed for 2 hrs at 550° C, if desired, to determine the organic and inorganic proportions of the TSS.

Invertebrate Analysis

Core samples were sorted in the field and preserved with formalin. In the lab, preserved invertebrate samples were transferred into a shallow plastic tray with a little DI water added. Invertebrates were removed by hand using a dissecting scope and/or large magnifying glass where needed. Invertebrates were stored in glass vials by higher taxon (e.g., polychaetes, amphipods, taniads, decapod-shrimp, decapod-crabs, etc) for later identification to genus and/or species level where practicable, or family where not. We discussed “problem species” with benthic colleagues to clarify identifications.

We obtained numbers of each taxonomic unit per area cored. Abundances were compared between controls and disturbed plots using simple ANOVA. We also used the vegan package in R to assess diversity and species similarity and dominance (the latter two using adonis function in vegan).

Analysis Methods

Data from experiments were first analyzed by individual experiment, and later analyzed collectively to derive summary statistics (mean, median, etc). Statistics were calculated using ANOVA and RStudio. Measurements of sediment plume characteristics from the shallow COM sites and the seagrass sites were analyzed for trends along distance gradients from the disturbance, and downstream measurements were collectively compared to upstream or Control tracks. At the seagrass site, measures were compared between treatment Tracks and along the distance gradient from the disturbance baseline. “Focused Comparisons” were also made between the tracks at only Zone “A” (50 ft). At the deep water COM site, results were compared between the locations of the 4 measuring stations (North, East, South, West). Results were summarized into a Summary Results Table.

Image Analysis of Epiphyte Accumulation

Scanned images were analyzed in the ENVI 5.0 program (L3 Harris Technologies, Ohio). This geospatial analytics software is widely used for earth science, construction, and vegetation research, including several seagrass mapping studies (Lyons et al., 2012). Here, it was applied on a micro-level to analyze epiphyte accumulation on the leaf surfaces. Manually-identified regions of leaf scans representing either epiphyte colonized areas, or uncolonized leaf areas, were used to build spectral profiles to train the software to classify the covered vs uncovered leaf areas. The area of each seagrass leaf and associated epiphytes falling into the two categories were quantified by counting the pixels of the uncovered and covered areas of the leaf, respectively. The proportional coverage of the seagrass leaf by epiphyte was considered to be an indicator of epiphyte recruitment and growth relative to seagrass leaf growth, and it varied with environmental conditions.

Analysis of UAV Imagery for Sediment Plume Characterization and Tracking

In RGB color imagery, sediment plumes generated by disturbance of the estuary bed, appeared as a grayish trail in the natural scene of the benthic environment (Figure 2).

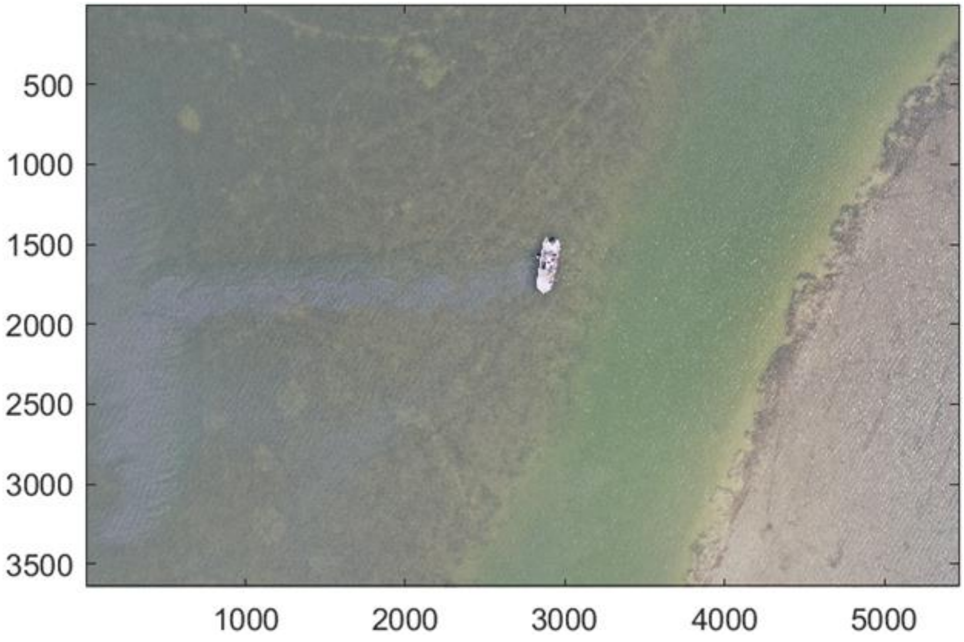


Figure 2. RGB color image

In order to characterize and track the plume and its temporal movement, the following preliminary image processing methods have been identified:

Step 1. Convert image into an alternate color representation, such as HSV. This color scheme increased contrast between the sediment plume and the background (Figure 3.)

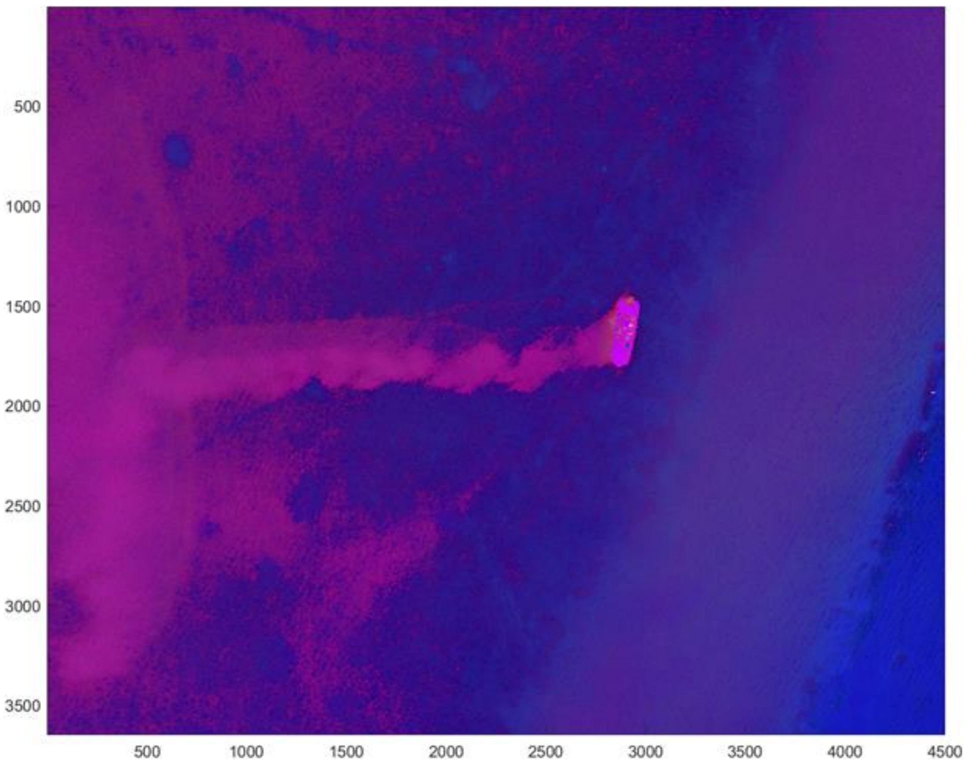


Figure 3. HSV color representation

Step 2. Frames were identified and their weighted combination that provide the highest contrast. Typical comparisons included difference over sum of two frames that provide the highest contrast.

Step 3. Identify the appropriate dynamic threshold and obtain the binary image representing the sediment plume. (Figure 4, left)

Step 4. A morphological image processing tool was applied to remove specular noise or unwanted pixels, such as erosion, dilation or opening, closing. (Figure 4, right. In this example, morphological opening was applied with a 3x3 cubic structuring element.

Figure 4. Binary image representing plume, with noisy pixels (left). Binary image representing sediment plume after noisy pixels were removed using morphological image processing operations.

Characterization of the plume: This was achieved based on the area covered by the plume. One method to do this was to determine the pixel size using a reference in the image. The length of the boat was used as a reference to determine the area of the plume as a rough measurement or starting point, since the size of the boat is known. A yard stick (ruler) was also placed on the boat and its tick marks used to calibrate the area each image pixel represents. This was possible where the UAS flew low enough with high resolution.

Alternatively, height of the UAS when the image was captured was used, along with camera properties, so that the area represented by each pixel could be calculated and total plume area reported.

As an example, in Figure 4 (right), the total number of pixels identified as plume was determined as 1,155,079 pixels. The size of the image shown was 3648x4500 pixels. The image had a total of 16,416,000. In this simple example, ignoring the pixels belonging to the boat, the plume cover of the captured image was ~7% of the natural scene.

Tracking the plume: Optical flow was used to track the temporal movement of the sediment plume. Optical flow determined the speed and direction of motion across two image frames acquired at different times. Alternatively, tools such as BACI were explored for comparisons of data as a time-series.

An example of optical flow is shown in Figure 5.

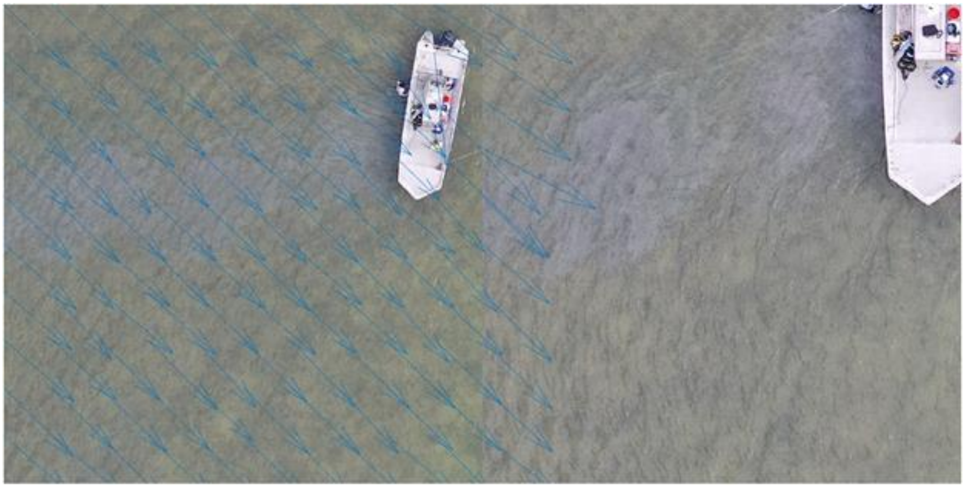


Figure 5. Optical flow for tracking sediment plume movement across image frames.

Invertebrate community structure analysis

We counted numbers of each taxonomic unit per area. Abundance was compared between controls and disturbed plots using simple ANOVA and linear regression techniques where tests of statistical assumptions warranted. Otherwise, generalized linear modeling was conducted testing against other distributions (I.e., Poisson and Negative Binomial).

We used the vegan package in R to assess diversity and species dissimilarity (the latter using adonis2 function in vegan).

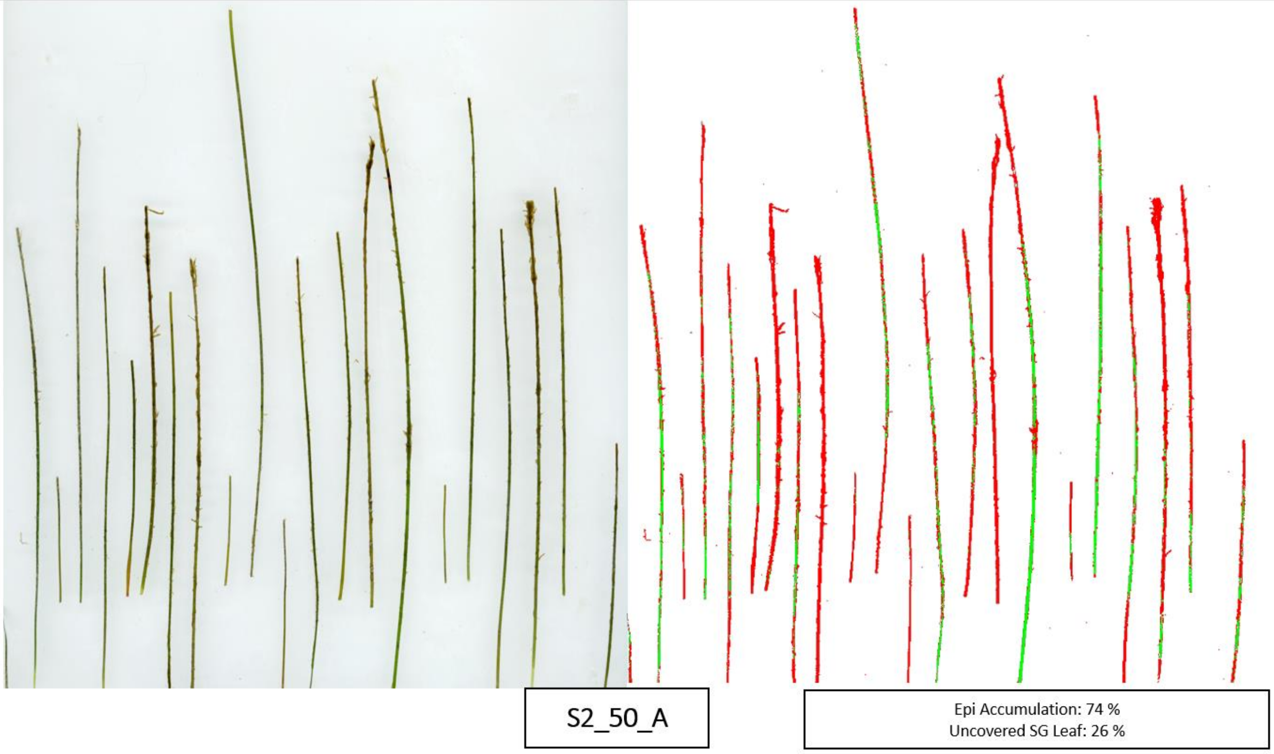
All response variables were tested for differences: a) between disturbance treatments (Tracks); and b) along gradients of distance downstream from disturbance source.

Analysis of Light Attenuation

Both light attenuation measurement methods were deployed in a few instances to confirm the correlation between the two measures. Dips in plots of available light over time indicated passing sediment plumes. *Relative Difference,* defined as the difference in light level between an upper logger and a lower logger, divided by the upper logger reading, provided a metric that corresponds to the percent of incident light at the top logger that is attenuated by the water column before it is perceived by the lower logger. Plots of *Relative Difference* show passing sediment plumes as an increase in the difference over time. Alternatively, sediment plumes could be detected as changes in the area beneath the curve representing the total light received. This allowed calculation of the total reduction of light experienced (light budget) during a sediment disturbance plume and indicated the persistence of the plume.

Seagrass Imaging and Analysis

Seagrass samples from each harvest were scanned for subsequent image analyses.



Samples of *Halodule Wrightii* were scanned on a white background with leaves organized by shoot (Left). Image analysis interpretation by ENVI is shown on the right, where green indicates seagrass leaf uncolonized by epiphytes, and red indicates epiphyte colonized areas. Pixel counting showed that of the total area of seagrass plus epiphytes, 74% was epiphyte and 26% seagrass surface.

To quantify epiphyte percent accumulation on the *Halodule Wrightii* blades in ENVI, similar pixels were grouped based on RGB values using the Color Raster Splice tool. Pixel values that were either seagrass, or epiphyte were saved and unnecessary parts of the image were removed (Resize Raster tool). A Region of Interest (ROI) was defined with the use of a threshold and resizing. For this picture a Band Ratio of Red/Green was used to distinguish pixels of epiphytes vs seagrass. The Pixel information Tool was used to determine a selective ratio value, in this case seagrass ratios fell in a range of 0.05 to 0.90, and epiphytes had values of 0.91 and above. Lastly, Raster Color Slice removed other classes (colors) and seagrass and epiphyte pixels were assigned green and red colors, respectively, and pixels of each were counted.

RESULTS

Overview, Challenges and Adaptations

The team's original plan to perform measurements at COM sites prior to initiating seagrass experiments had to be adapted because of difficulties in arranging field work. Issues such as boat availability, different researcher and student schedules, Covid infections among students and weather delays set back our efforts to scout seagrass sites and access COM operations. Although these problems eased somewhat, they persisted to some degree through summer and fall (2022) with the end result of making fewer field work trips than planned. We adapted by initiating seagrass site work while we began to collect sediment disturbance data at COM sites. This dual emphasis went on through the project end.

Generating sediment plumes at monotypic, dense stands of *Halodule wrightii* seagrass was complicated hydrologically. The assumptions that current would be significantly wind-direction associated and that tidal forcing would come from the nearest deeper water direction proved false. Turtle Bayou, a muddy seagrass site, became untenable after numerous attempts to generate predictable sediment plumes failed because of the unexpected hydrology of the site. Efforts remained focused on the relatively sandy San Jose seagrass site. Though similar hydrological complications arose there as well, we successfully adapted our sediment plume generation methodology in response to the water currents. Examples of these issues are illustrated below. Adaptations included re-orienting the disturbance location and measuring stations. Hydrology was variable on short timescales (down to minutes) and proved the major challenge. Drone (UAS) imagery became a critically important tool in our adaptation efforts, but availability was limited by the aforementioned factors.

Sediment plume measurements, described in greater detail below, began in summer 2022 after some initial instrument field tests in the spring. We began efforts with wadable shallow water research COM operations, but the two Harte Research Institute (HRI) sites in Copano and Palacios were winding down operations (ending in June and August, respectively) and only a small number of oyster cages remained. This meant that there was limited operational activity to observe. A third, but smaller COM research site (Hollenbeck; <15 cages) initiated in spring 2022 at the TAMU-CC campus, was also utilized for measurements. After initial observations, we extended our work by simulating disturbances at each site by having students walk around the cage lines similar to how operators might work. The shallow water COM sites had high wave energy and ambient turbidity, complicating instrument deployments and sediment plume observation/measurement. Deep water COM measurements were made at the TX Oyster Ranch (Copano Bay, Mr. Brad Lomax) from October through March, and made use of a buoy-suspended wire cage to deploy instruments. Again, the UAS imagery proved very informative as it provided a view of where, and where not, the sediment plumes traveled, as well as estimates of their rate and distance of travel.

Conditions and Sediment Plume Measurements at COM Sites

COM Disturbance Log and Summary

A detailed disturbance log recorded metadata such as disturbance parameters (p-m) and associated weather and physical conditions, for each experiment, to facilitate data analyses. This log is a large spreadsheet which is separately uploaded to the GLO.

Various visualizations and measurements are presented below for sediment plumes at both shallow and deep COM sites. Data are reviewed by technique, eg. visualizations, light logger data, TSS and % organic content, and settled sediments (traps) and % organic content. These data are summarized and analyzed collectively at the end of the Results section.

Visual Observations of Sediment Disturbances (Pictures and Video Captures)

Shallow water COM sites were visited July through September to perform disturbance measurements. On several occasions, measurements and video were recorded at the Palacios HRI site while the COM operator worked. Copano HRI had the last of its cages removed, Palacios was down to a single line of cages, and Hollenbeck farm was starting up with only ~10 cages by August. Artificial disturbances at each site, caused by researchers walking around the cage lines as though tending cages, were used to simulate sediment disturbances to accommodate additional measurements. These artificial disturbances, usually made by 2-3 persons walking around the cageline for 5 – 30 min, would probably represent the walking aspects of COM operation, but not the sediment clouds released from the cages during bag shuffling or harvesting activities (see later).

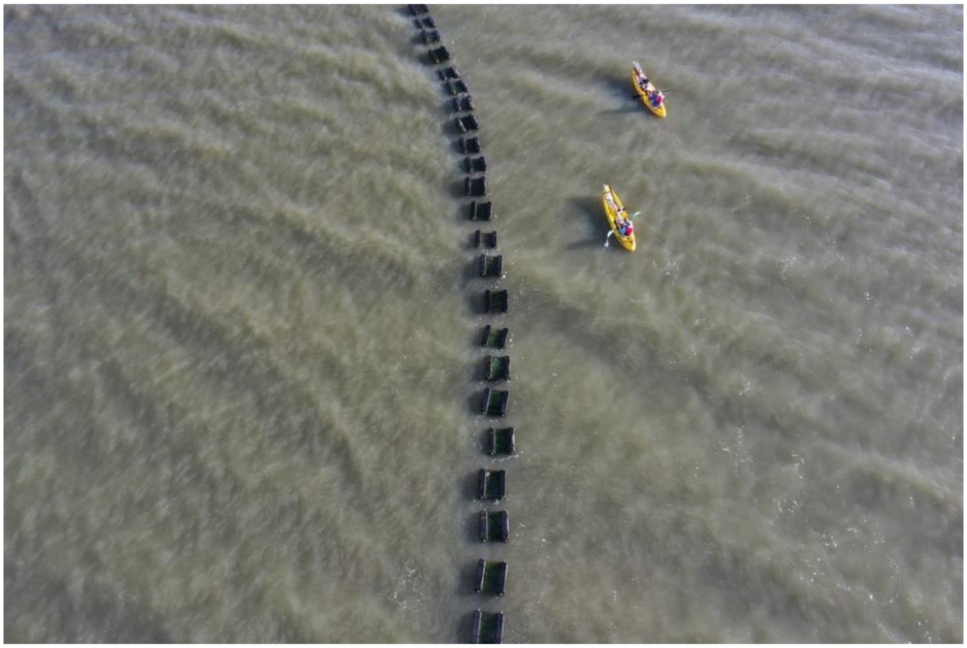
Copano HRI Site

Deploying Light Loggers and Sediment Traps down-current of the cage line

3 lines of measuring poles (Light Loggers and Sediment Traps) down-current of the cage line at Copano HRI

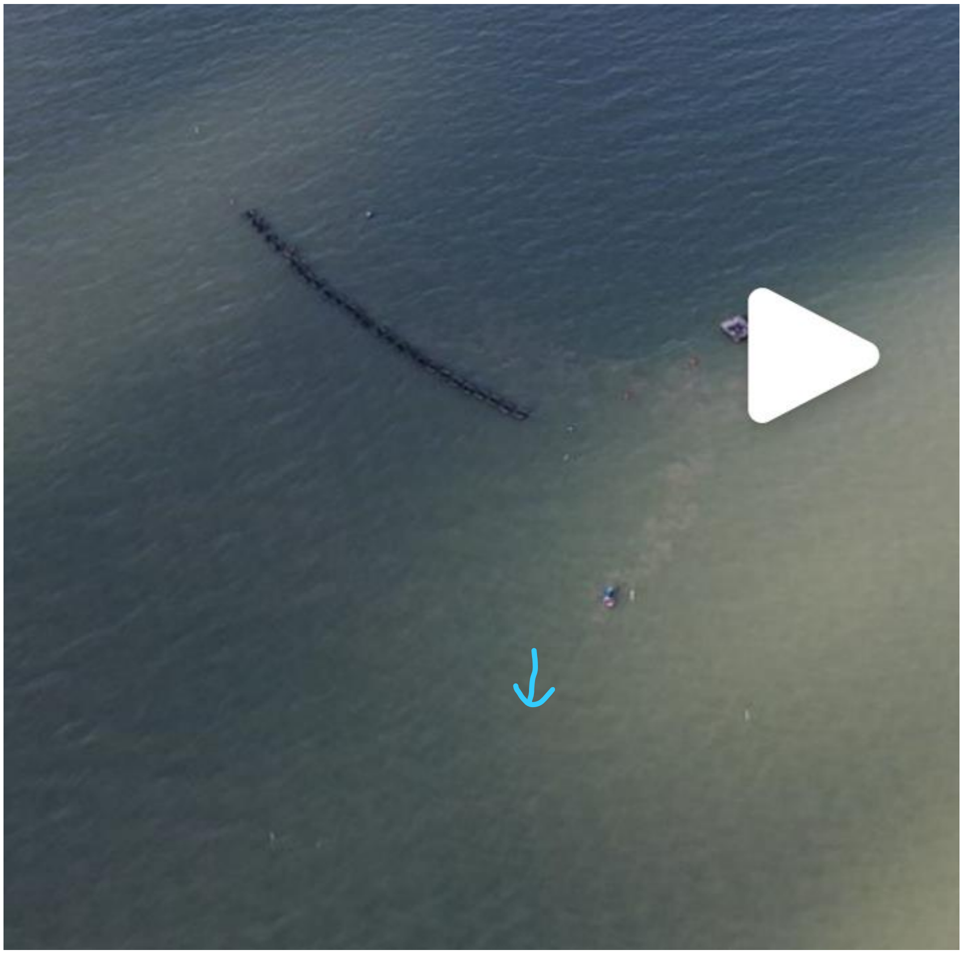
Recovering measurement equipment following the disturbance

Palacios HRI Site (Still active at the time)



Team members going out to deploy measurement gear. Positioning the poles to be down-current of the cage line was difficult in the changing conditions. Note angle of waves to cageline and the siting decisions required to place measurement stations at different distances down-current. On multiple occasions, the current would shift during the ~ 1 hr required to setup the measuring stations, requiring a new setup before undertaking the experiments.

Palacios COM operator tending cages. He is switching oyster bag positions within cages and switching some into different cages (shuffling).



Research team member retrieving sediment traps and TSS samples following a simulated disturbance experiment at the Palacios COM. Note the sediment plume generated by walking to the measuring station. It is visible in the already turbid water. Sampling each station without introducing additional disturbance to down-current stations was difficult. Harvesting samples from the farthest, most down-current stations resulted in different sampling times at each station.

Hollenbeck (TAMU-CC)

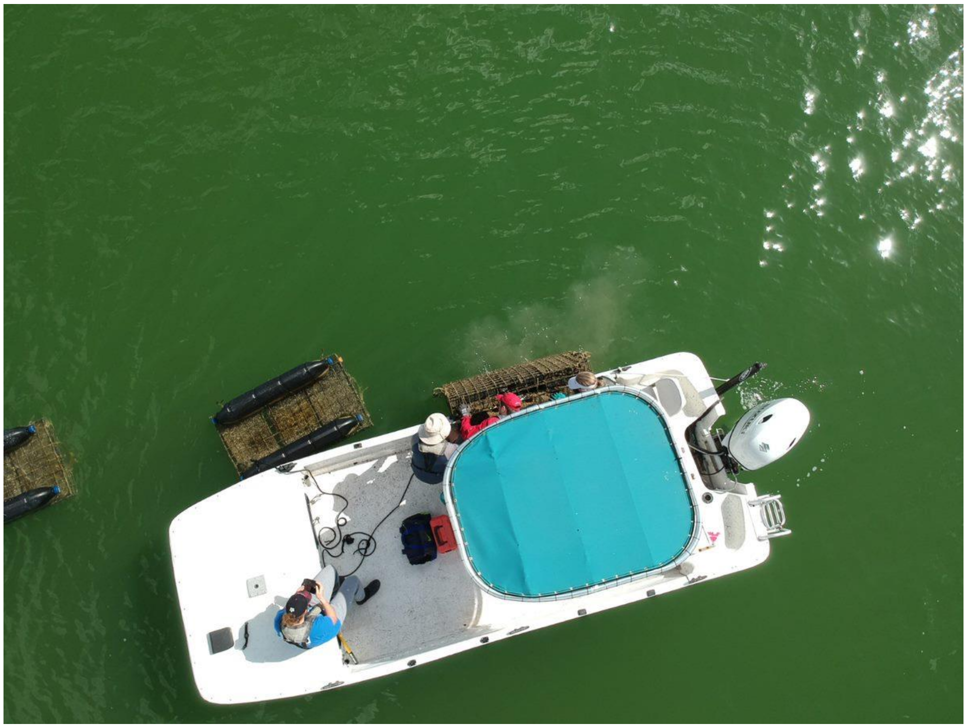
The Hollenbeck Corpus Christi Bay COM is a small operation (< 15 cages) that is easily accessible. Land and a breakwater immediately adjacent on two sides create eddy currents, and the bottom is uneven (alternating guts and bars) due to wave energy. The arrow in photo below indicates the current direction, perpendicular to the waves. Team members are retrieving samples from 3 lines of measuring poles.

Lomax COM Site (Copano Bay)

The Lomax COM site (TX Oyster Ranch) is currently a mid scale commercial oyster mariculture facility utilizing an off bottom, suspended cage form of mariculture. It has an average depth of 8-10 feet. This site is exposed to current from different directions depending on weather, with the predominant current leading in a Northwesterly direction from the prevailing SSE winds. Aerial pictures below show the layout of the farm, some operational activities, and placement of a floating measurement platform.

Overview of TX Oyster Ranch (Lomax farm). Red arrows indicate our measuring stations

Shuffling bags within cages. Note small plume of material released from bags and cages when flipped back into water following handling.



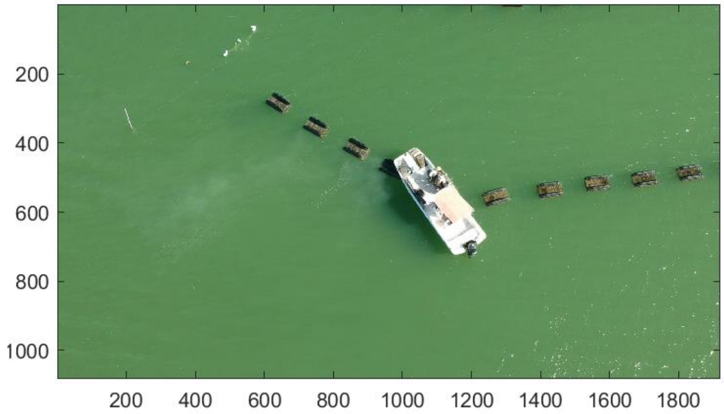
Shuffling bags within cages. Note small plumes of sediment.

Reloading bags of tumbled oysters back into a clean cage.

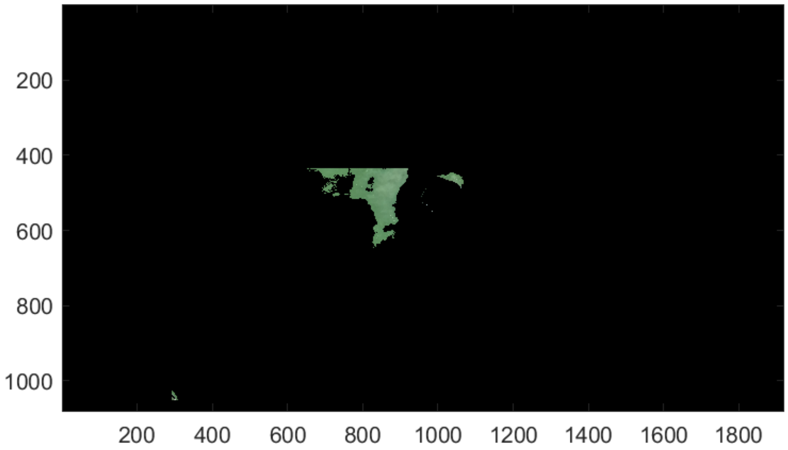
Measuring station at Lomax farm under calm conditions.

Image Analysis of Sediment Plume Movements at COM Sites

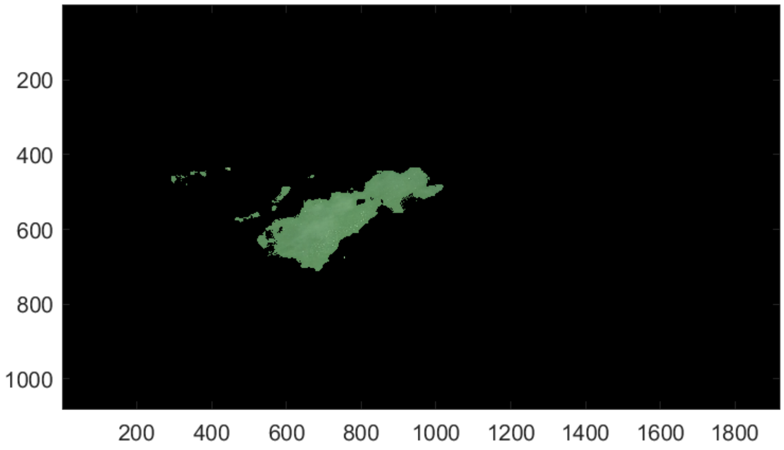
Original and MatLab-interpreted image series of a small sediment cloud generated at a deep water COM site (Lomax farm) upon routine oyster cage maintenance (flipping cages and shuffling bag positions in this case). Video indicates the small plume dissipates within a short distance and time. Sediment plumes coming off the cages are hypothesized to be primarily biodeposit and organic material.



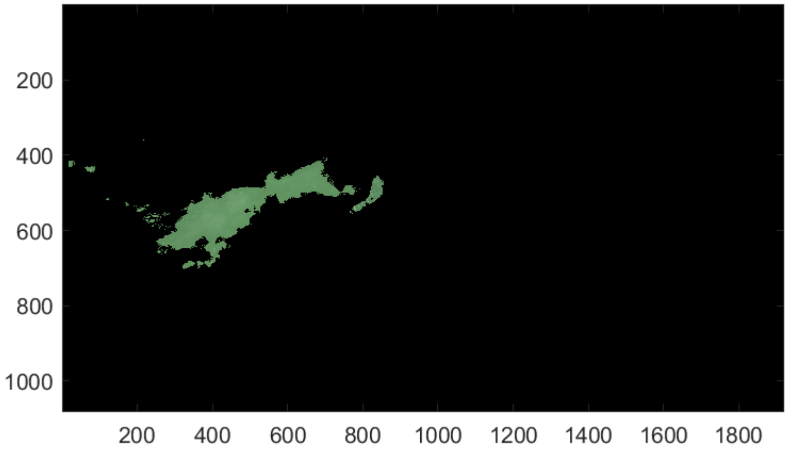
The 3 extracted video frames (above) were filtered 8 times with 5X5 averaging filter and a small rectangular Region of Interest (ROI) was selected and remainder of image was masked. The image was converted to HSV color space, and thresholding parameters were identified. After that, the image was thresholded to obtain the binary mask representing plume within the ROI. The binary mask was overlaid with the original image for a visual of the plume area.



Percent plume of rectangular ROI for frame 105 sec (1:45 into the video): 4.36%



Percent plume of rectangular ROI for frame 195 sec (3:15 into the video): 10.97%



Percent plume of rectangular ROI for frame 225 sec (3:45 into the video): 10.65%

Light Logger Data

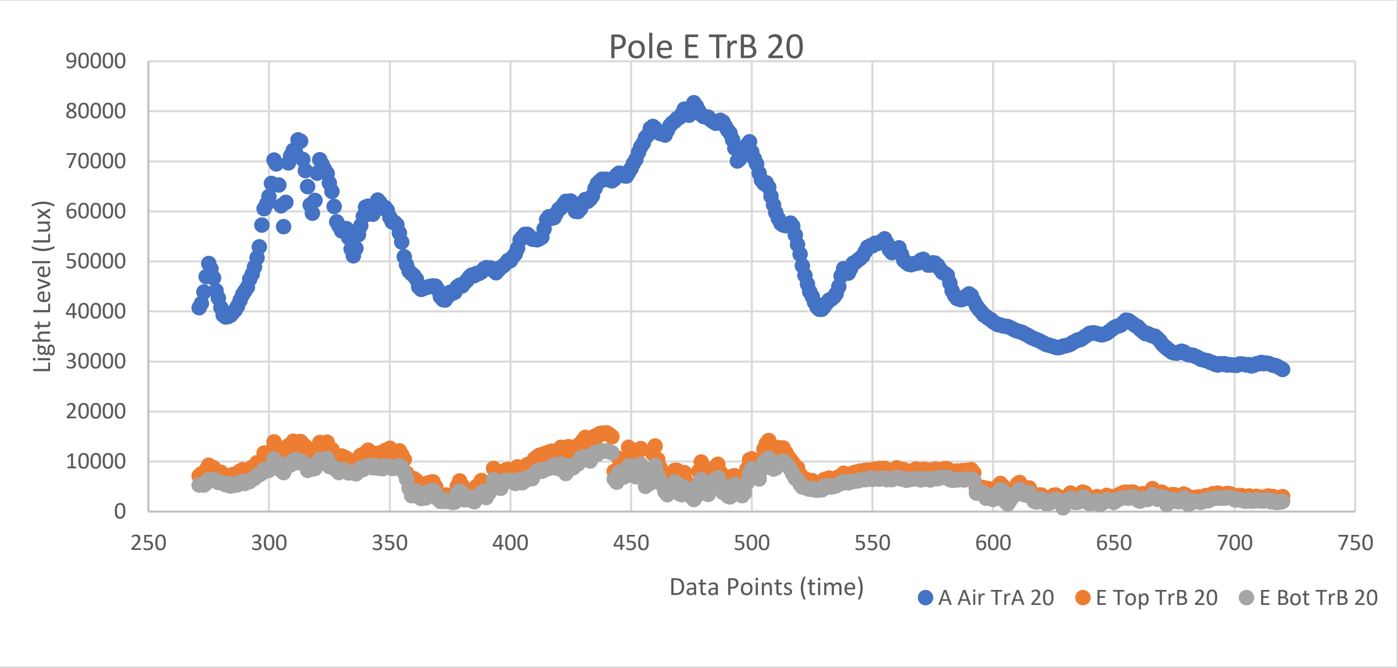
Results from Shallow Water COM sites

Examples of Light Logger measurements from each shallow water COM site during a simulated sediment disturbance, are shown below. Because of the shifting current directions, it was best to deploy 2 or 3 lines ("Tracks") of light loggers in radiating directions to assure capturing sediment plume movements. (Examples of this are illustrated below)

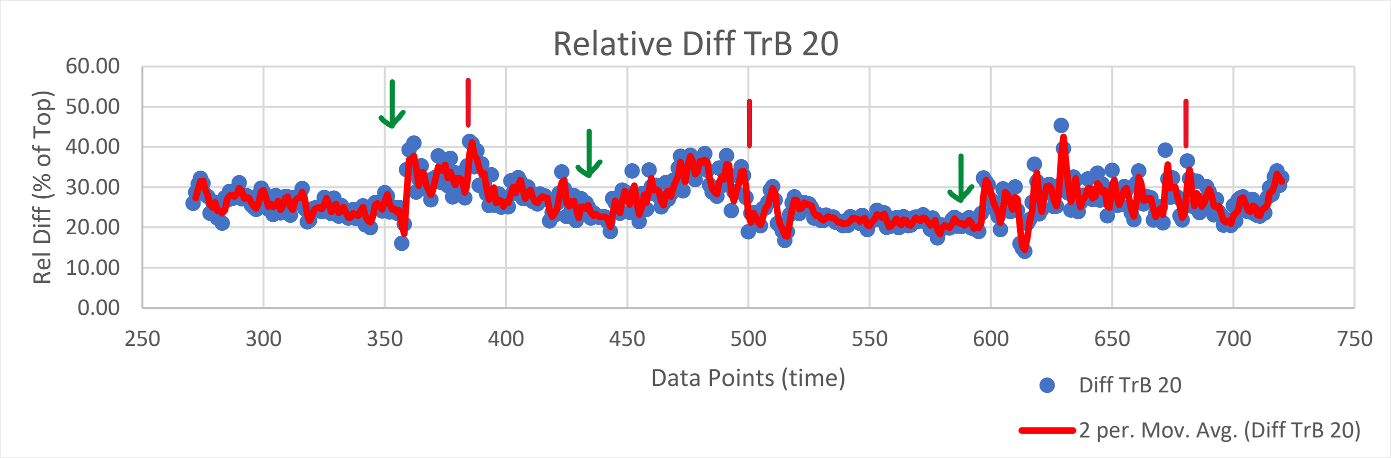
Light Logger traces record ambient light with an "Air" logger, and 2 loggers mounted at different depths in the water column at an approximate position of where a seagrass canopy would be located. The difference in light detected by the two loggers provides information on the attenuation of light by turbidity in the water column. The difference, expressed as a percent of the reading at the upper logger, provides "*Relative Difference*", a value that will increase with the passing of a sediment plume. Typically multiple sediment disturbances are applied with disturbance intensity measured as "Person-Minutes" (P-M).

Copano 8-31-22: Cloudy/stormy day; East wind & current well-aligned w/cage-line

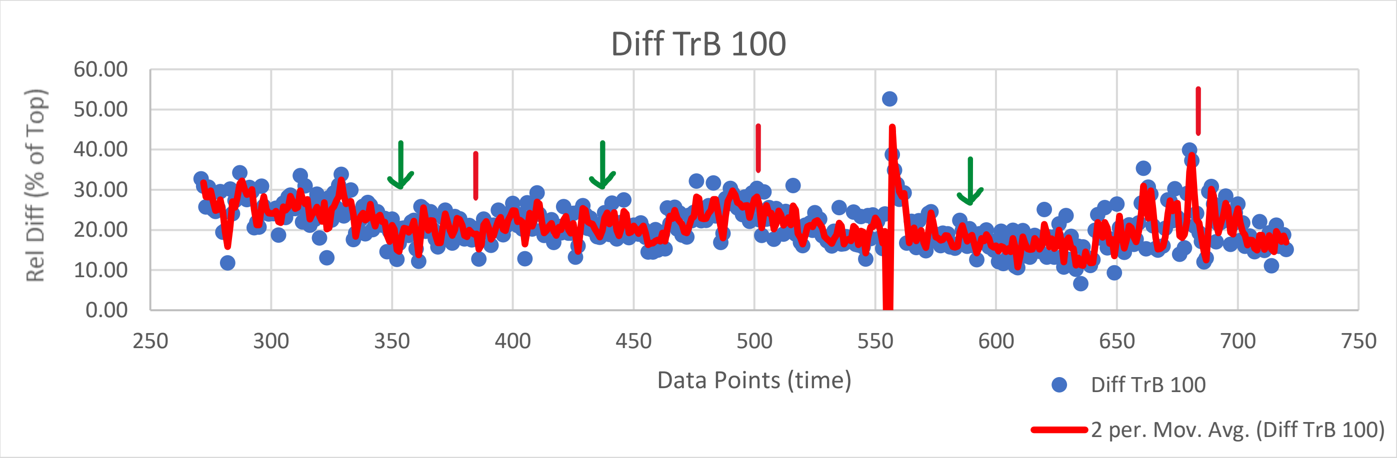
3 Tracks of poles (A – C) and 3 Disturbances: I – 20 p-m 3:57-4:07 (352-384); II – 80 p-m, 4:26-4:46 (439-501); III – 150 p-m, 5:16-5:46 (589-681) **[Green=Start; Red= Stop]**



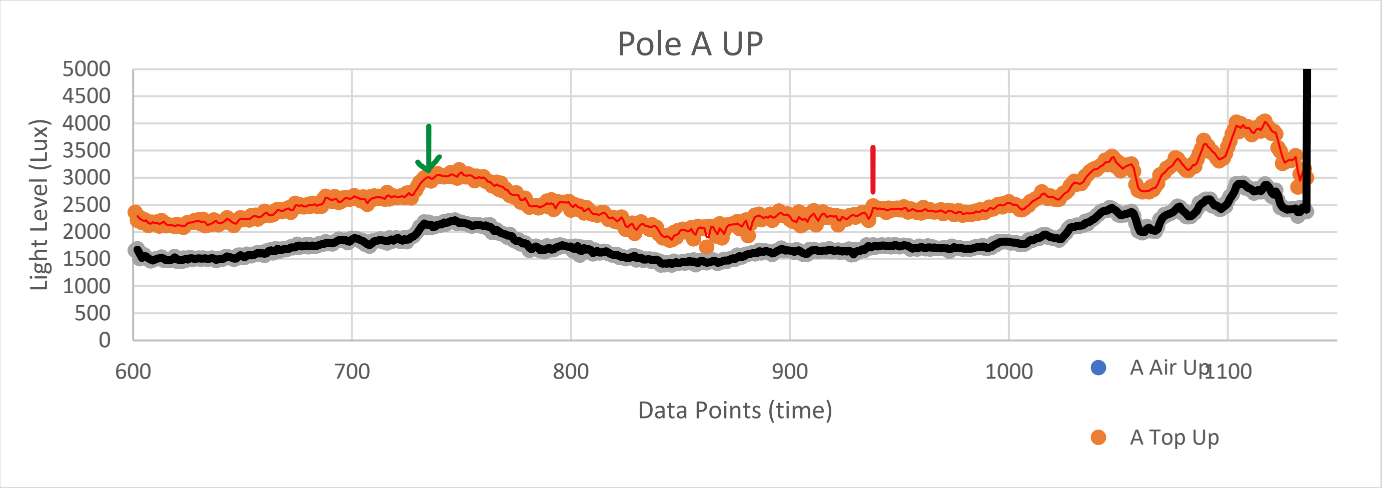
Relative Diff = Difference between Top & Bottom Loggers *RELATIVE TO* Top Logger (x100) [= Diff as percent of Top] at Track B 20 ft from the disturbance baseline (source).



Note that the baseline Relative Difference increases shortly after initiating a disturbance 20 ft away. Light traces become noisy and may also show "pulses", interpreted as waves of sediment plumes generated by passing walkers. The disturbance plume is harder to discern at 100 ft from the source, but it is noticeable towards the end of the disturbance (the delay in arriving at the logger pole provides information on its rate of movement).



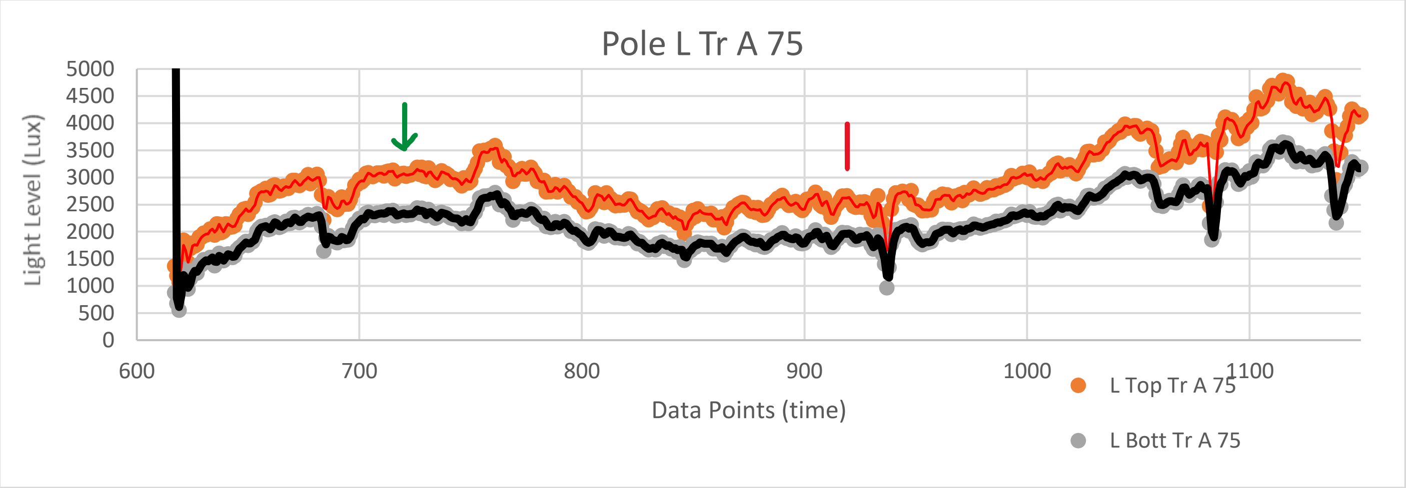
*Palacios HRI COM (8-24-22)***)** East wind; Outgoing tide; Together made strong current traveling perpendicular to oyster cage line; 60 p-m (pts 721 – 906)



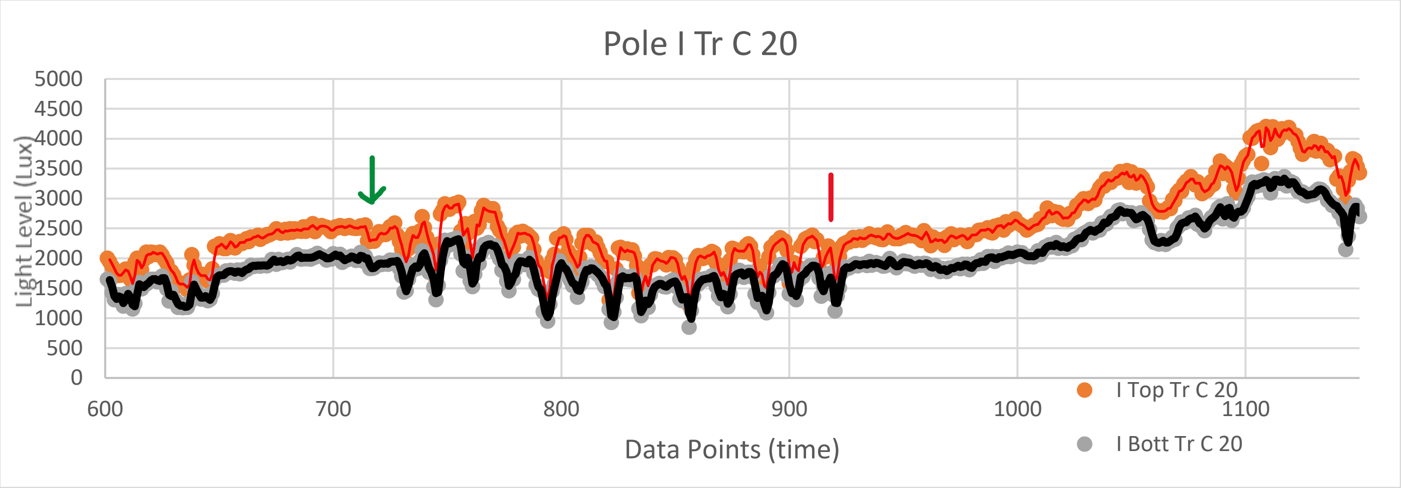
The up-current pole (above) shows no sign of sediment plume (applied from green arrow to red line).

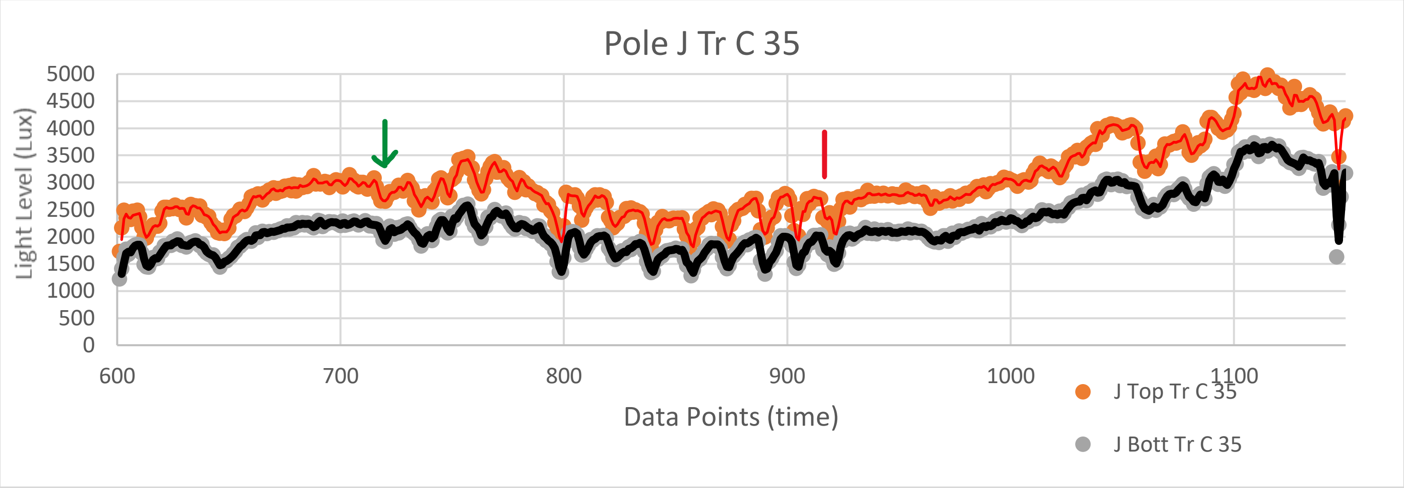
In contrast, down-current measurements (below) show strong, pulsating light level dips indicating the plume traveled over 75 ft along both Tracks A and C. Time of sediment plume arrival at each pole can be discerned.

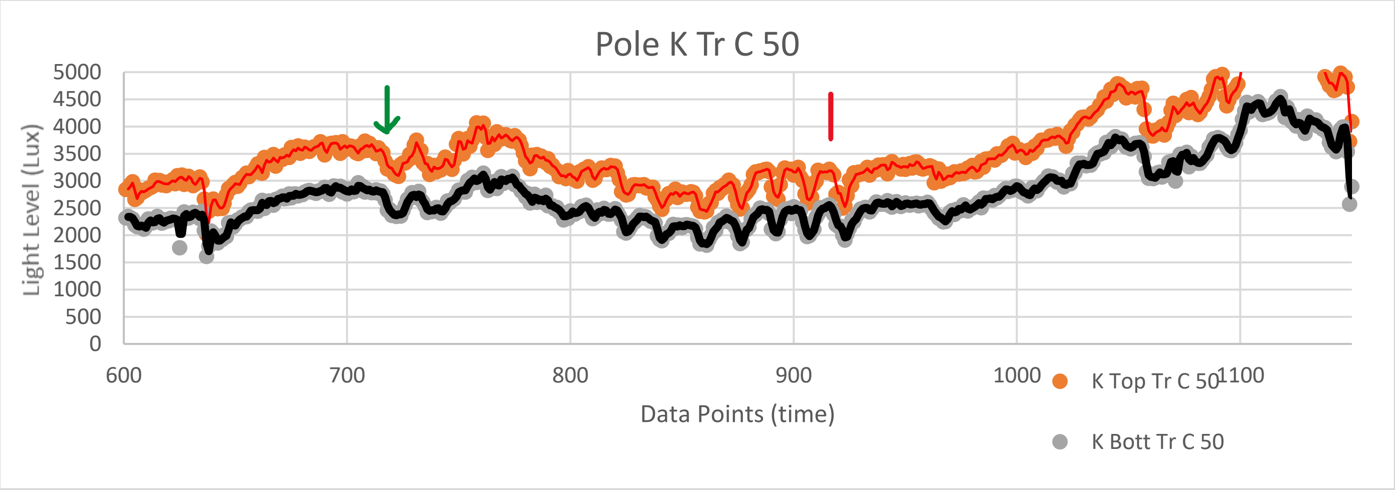
Disturbance weakest along Track A, but still reached 75 ft pole



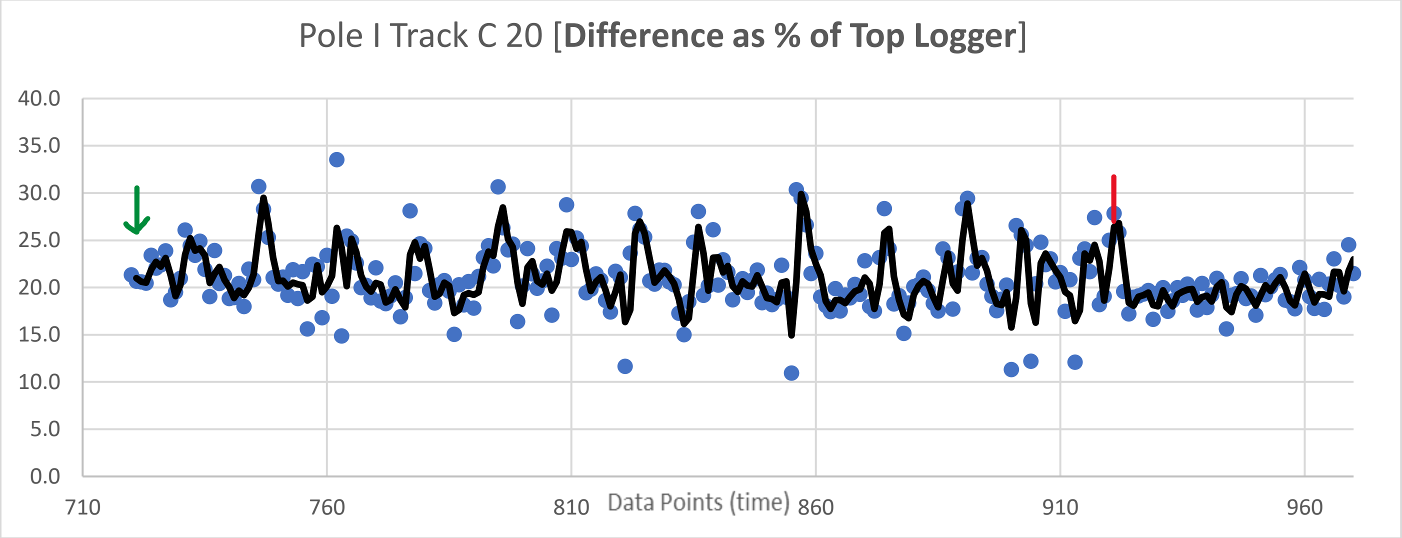
Disturbance strongest along Track C







Relative Difference Analysis (Difference between Upper & Lower Lt Loggers at 20 ft): Peaks represent increased light attenuation (ie pulsed sediment plumes) upon up-current passing of disturbance walkers. Expressed as % of upper logger reading. (Zoomed in on one disturbance time period; Black line represents a 6-point moving average to smooth).

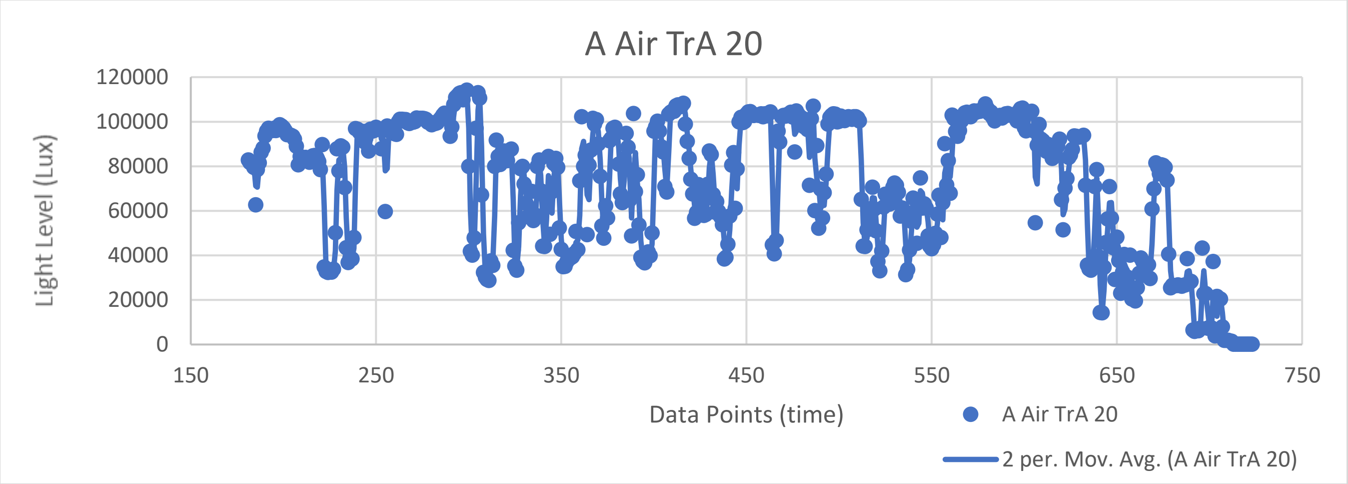


The baseline attenuation of light is approximately 20% (for 13 cm of water column between the two light loggers). The pulsating peaks indicate sediment plumes generated by the walkers, and attenuation increases to 30% or greater. Thus, the attenuation due to generated sediment plumes is approximately 10% in this measurement.

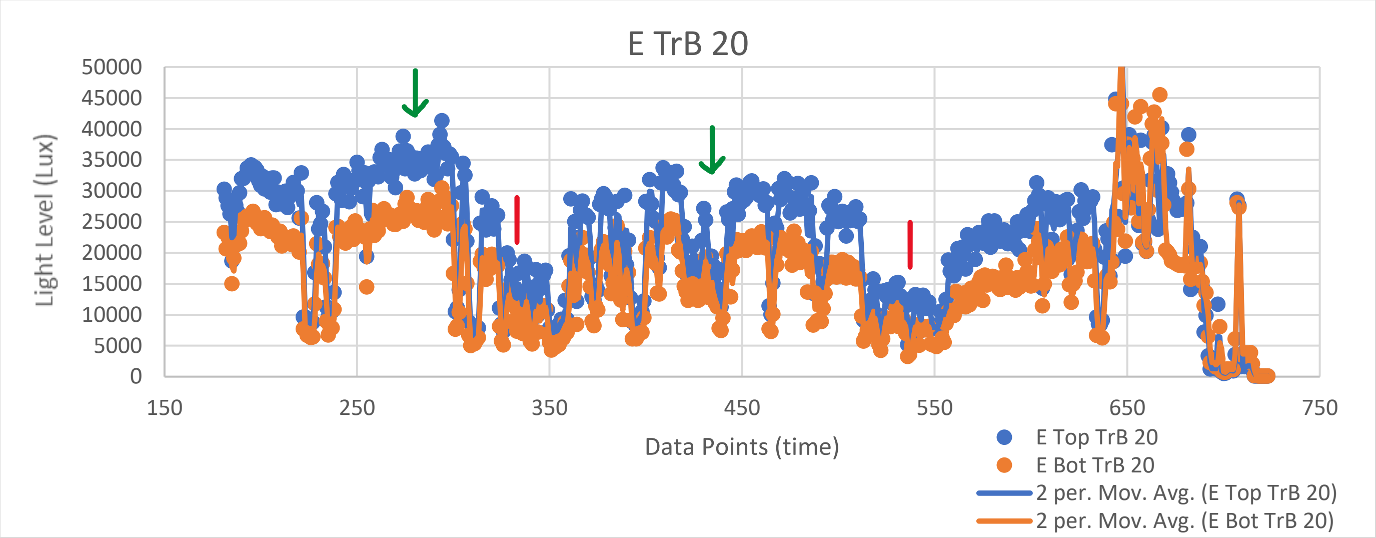
Hollenbeck (9-2-22) 2 Disturbances, Weather conditions: Light wind out of southeast around 3.7 m/s, mostly sunny with some passing clouds. Current is variable due to effects of jetty structure.

Dist 1: 277-324 (1:32:1:47 30 p-m) ; Dist 2: 438-528 (2:25-2:55 90 p-m)

[G=Start; R= Stop]

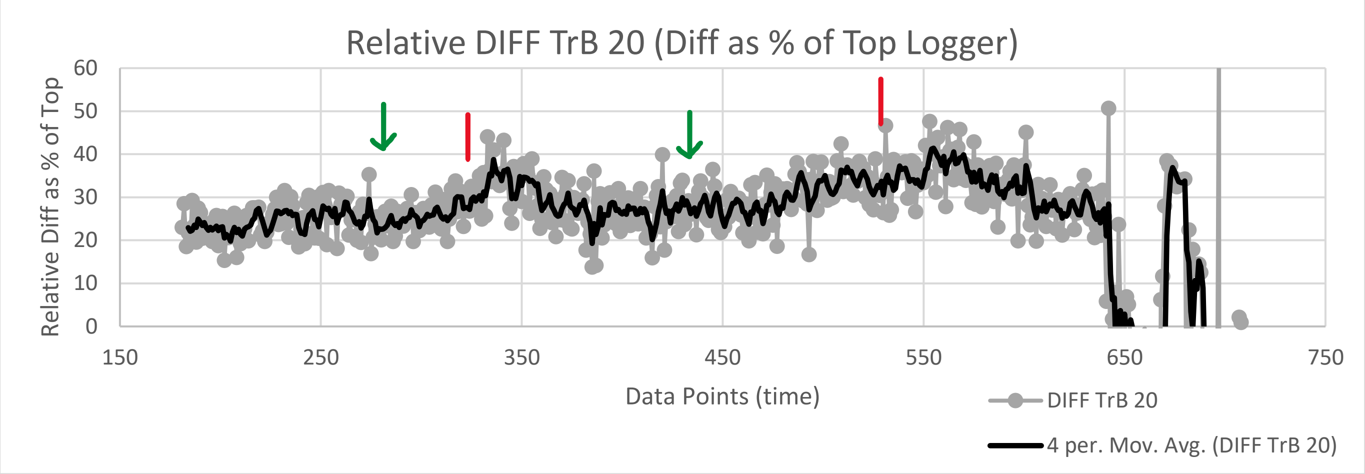


Air logger was very noisy which is not typical; Combination of partly cloudy and perhaps vibration in wave currents. Similar noise at both loggers on Track B pole at 20 ft.



Impossible to discern disturbance difference here just viewing light levels.

Relative Difference: Reduces the spiking noise to reveal attenuation increases of about 10% relative light difference with generated sediment plumes.



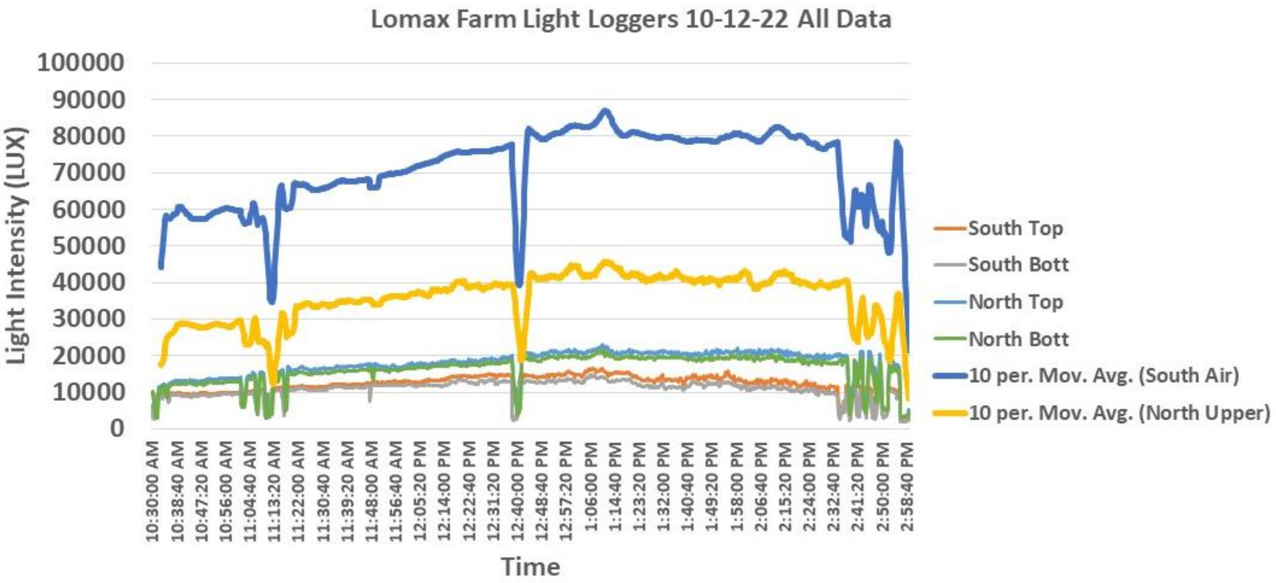
The difference measure reduces much of the noise and reveals ~ 10% - 15% attenuation increase with disturbance, as well as a return to the baseline attenuation after the disturbance. The larger 2nd disturbance (90 p-m) showed a greater attenuation, though not proportionately so.

Results from Deep Water COM sites

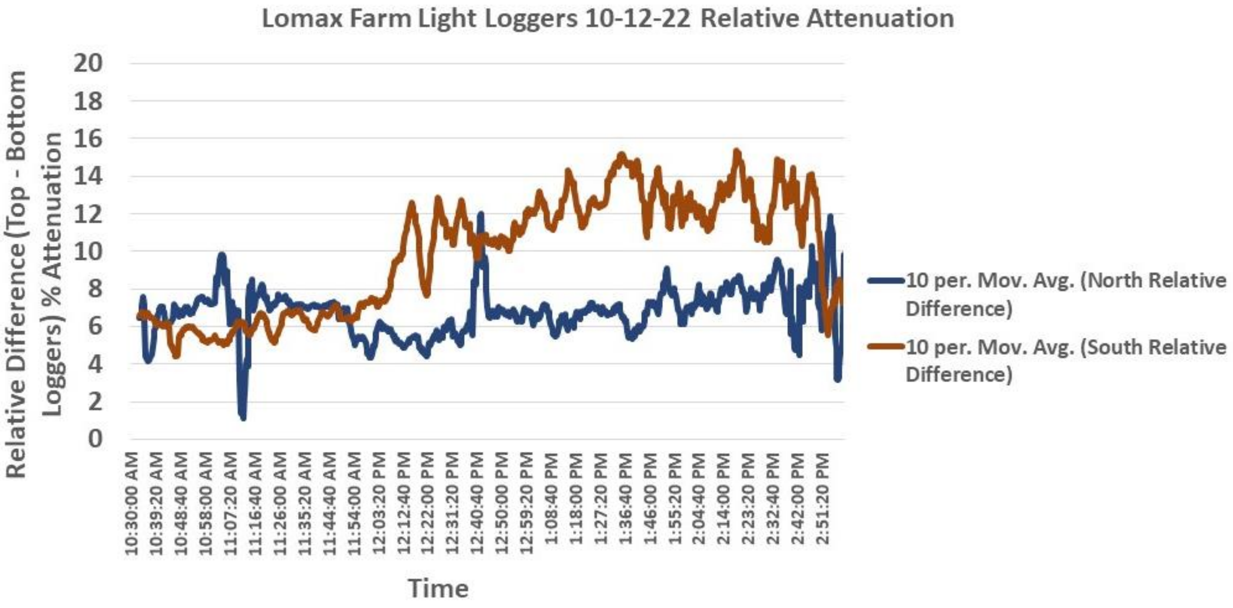
Measuring stations were deployed 5 times at the Lomax Copano COM site. Each station had 3 light loggers at different depths ranging from 1-4 ft, and a sediment trap. These were typically deployed in 4 sets, 1 each on the North, South, East and West sides of 7 active cage lines. In some cases, the instruments were very close to the cage lines (< 20 ft), and at other times they were located approximately 50 ft from the edges of the farm area (up to 100 ft from the cages). Deployments lasted from several hours up to 4 days. COM personnel were working what are considered typical maintenance actions, including cage flipping, bag shuffling, and harvesting on each of the days of deployment.

Lomax Deep Water COM (10-12-22) 1 day; Gentle S wind <5mph at 10:30 AM; rising to 4.1 m/s 135° SE at 1:45 PM; E 3.8 m/s at 3 PM, Sunny

Light loggers were setup with an "Air" logger above surface, an "Upper" logger at ~ 2ft deep, and a pair of loggers ~4-5 ft deep and separated by 13 cm (Top and Bottom, respectively).



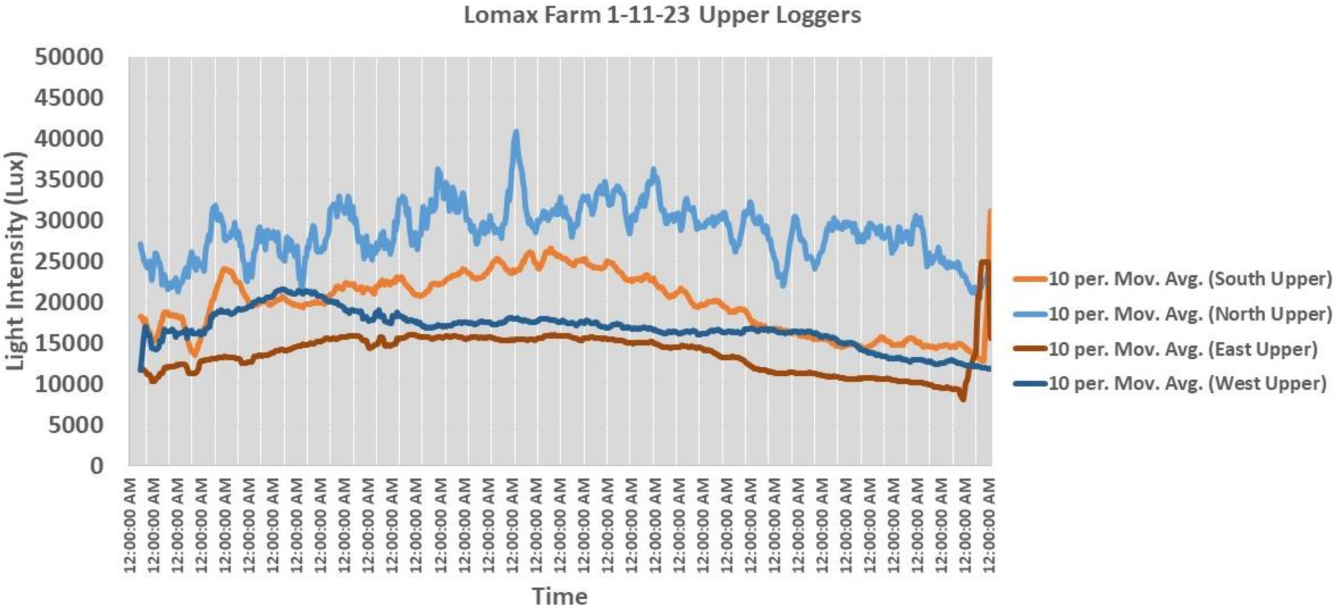
Relative Difference between Top and Bottom loggers increased with wind in afternoon.

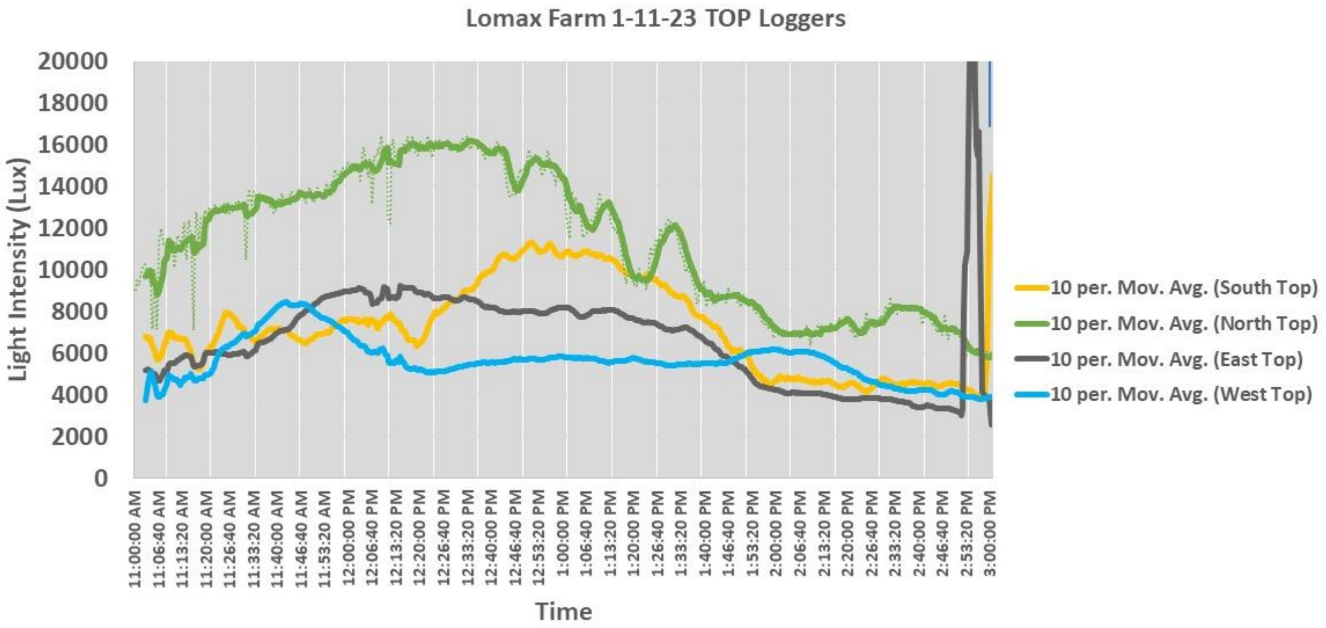


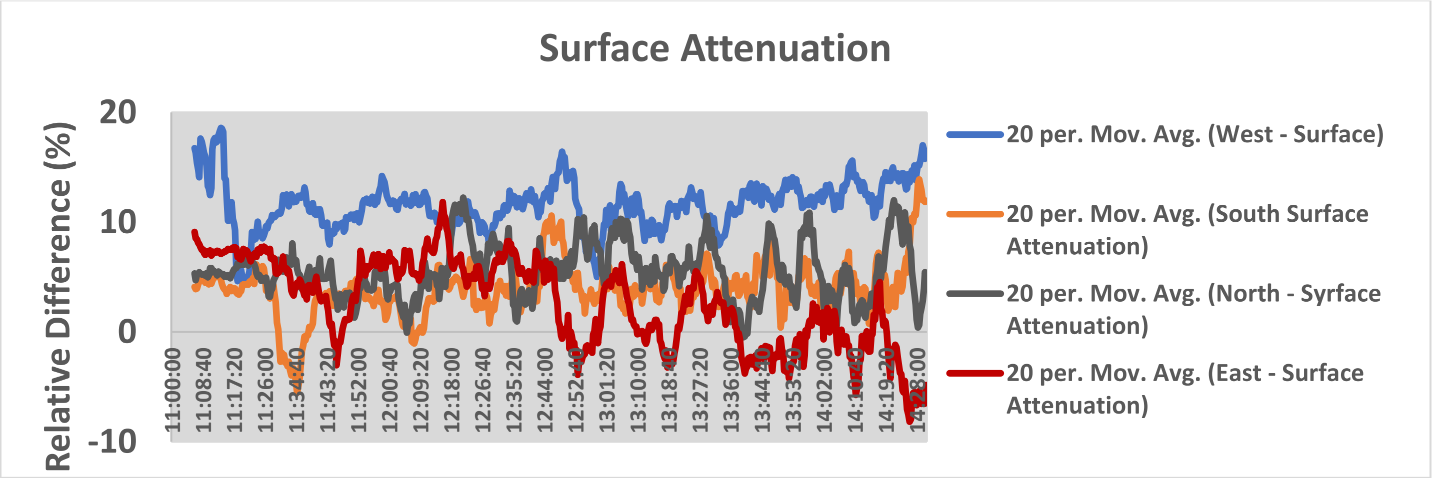
South logger (brown) was down-current; No obvious indications of oyster work-related disturbances. Attenuation increased from ~ 6% to ~14% with afternoon wind, so maximum difference in attenuation was ~ 8%.

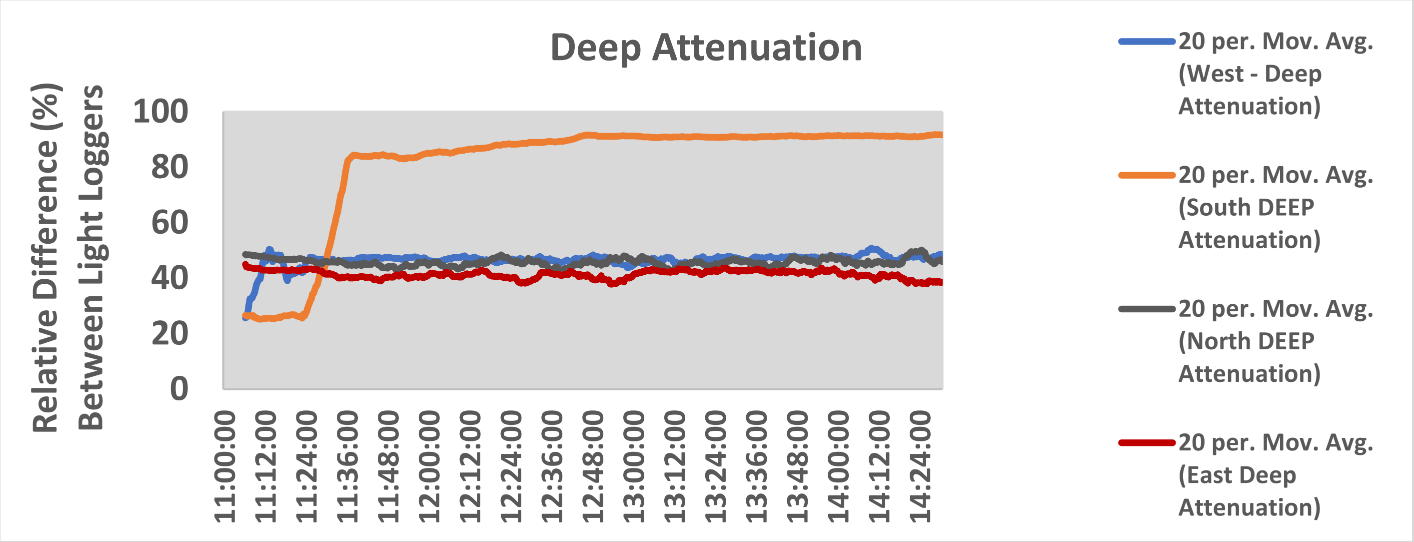
Lomax deep water COM (1-11-23) 1 day; Wind out of the South (180°) at 6.5 - 6.6 m/s (~15 mph) through the day. Sunny conditions persisted throughout the day with a few clouds. Regular COM operations conducted by farm operators during sampling period.

Light loggers were setup with an "Upper" logger at ~ 2ft deep, and a pair of loggers ~4-5 ft deep and separated by 12 cm (Top and Bottom, respectively).



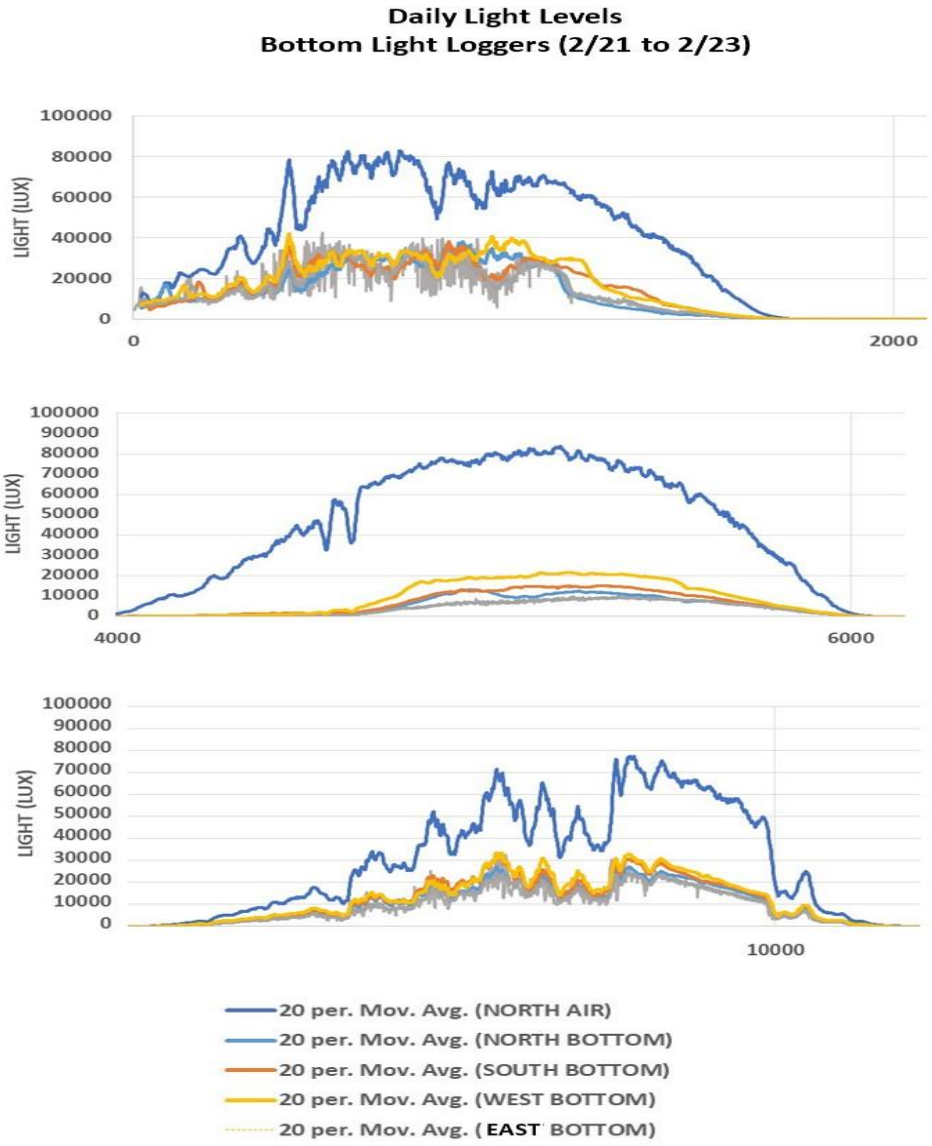

Lomax Farm 1-11-23 TOP Loggers
Light Intensity (Lux)
20000
18000
16000
14000
12000
10000
8000
6000
4000
2000
0
11:00:00 AM
11:06:40 AM
11:13:20 AM
11:20:00 AM
11:26:40 AM
11:33:20 AM
11:40:00 AM
11:46:40 AM
11:53:20 AM
12:00:00 PM
12:06:40 PM
12:13:20 PM
12:20:00 PM
12:26:40 PM
12:33:20 PM
12:40:00 PM
12:46:40 PM
12:53:20 PM
1:00:00 PM
1:06:40 PM
1:13:20 PM
1:20:00 PM
1:26:40 PM
1:33:20 PM
1:40:00 PM
1:46:40 PM
1:53:20 PM
2:00:00 PM
2:06:40 PM
2:13:20 PM
2:20:00 PM
2:26:40 PM
2:33:20 PM
2:40:00 PM
2:46:40 PM
2:53:20 PM
3:00:00 PM
Time
10 per. Mov. Avg. (South Top)
10 per. Mov. Avg. (North Top)
10 per. Mov. Avg. (East Top)
10 per. Mov. Avg. (West Top)

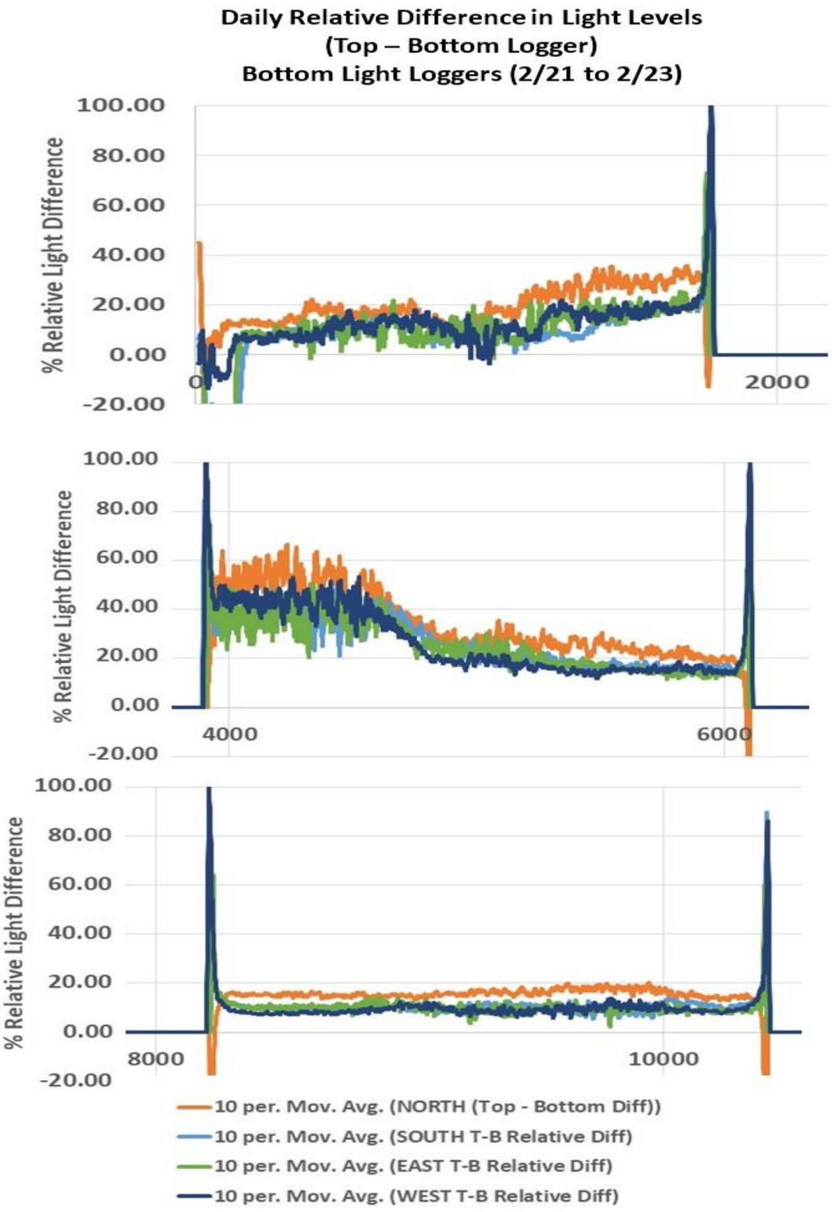

Surface Attenuation
Relative Difference (%)
20
10
0
-10
11:00:00
11:08:40
11:17:20
11:26:00
11:34:40
11:43:20
11:52:00
12:00:40
12:09:20
12:18:00
12:26:40
12:35:20
12:44:00
12:52:40
13:01:20
13:10:00
13:18:40
13:27:20
13:36:00
13:44:40
13:53:20
14:02:00
14:10:40
14:19:20
14:28:00
20 per. Mov. Avg. (West - Surface)
20 per. Mov. Avg. (South Surface Attenuation)
20 per. Mov. Avg. (North - Syrface Attenuation)
20 per. Mov. Avg. (East - Surface Attenuation)



Relative Differences between different measuring stations and/or times during day with activity less than 15% near surface and 45% at ~ 5 ft depth; No obvious indications of work-related disturbances can be discerned, but two of our loggers malfunctioned, and the South deep logger appeared to be in the soft bottom mud.

Lomax deep water COM (2-21 to 2-23-23) 3 day deployment; Weather conditions remained cloudy and foggy in the mornings, clearing up throughout the day each day during deployment. Wind shifted from SSE on 2/21, to S on 2/22, and then E on 2/23, and E to ENE on 2/24 (Day of pickup).
Wind shifted and caused greatest turbidity on 2nd day
Relative Differences varied from 5-30% on 1st day, 5-15% on the 2nd day, and 10-20% on the 3rd day
No obvious indications of work-related disturbances, but North side had consistently greatest attenuation.


Daily Light Levels
Bottom Light Loggers (2/21 to 2/23)
LIGHT (LUX)
100000
80000
60000
40000
20000
0
0
2000
LIGHT (LUX)
100000
90000
80000
70000
60000
50000
40000
30000
20000
10000
0
4000
6000
LIGHT (LUX)
100000
90000
80000
70000
60000
50000
40000
30000
20000
10000
0
10000
20 per. Mov. Avg. (NORTH AIR)
20 per. Mov. Avg. (NORTH BOTTOM)
20 per. Mov. Avg. (SOUTH BOTTOM)
20 per. Mov. Avg. (WEST BOTTOM)
20 per. Mov. Avg. (EAST BOTTOM)



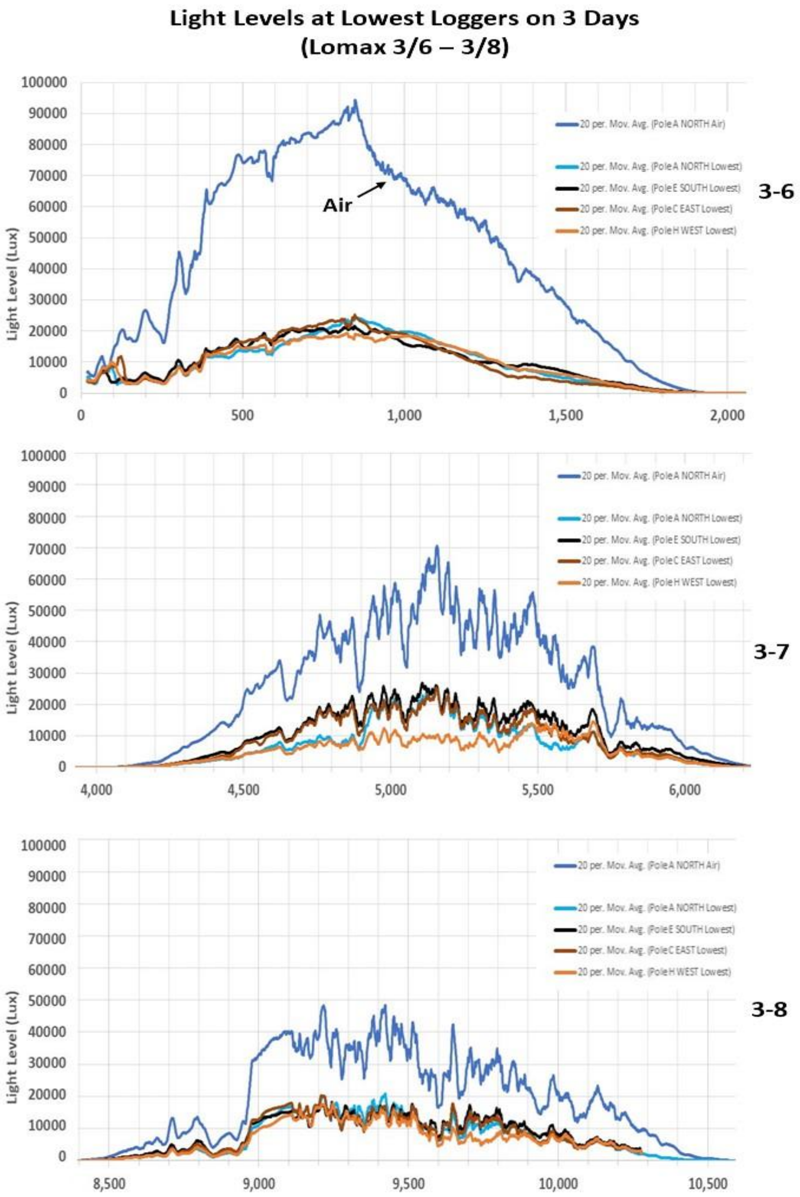
There were minor differences between measuring stations that arose during course of observations. These may or may not be related to activity at the farm, biodeposits from oysters, eddy-currents at the farm causing settling of suspended sediments in ambient current flow, or differences in bottom sediments being suspended by incident currents.

Lomax Deep Water *COM (3-6 to 3-8-23)* 3-day deployment;

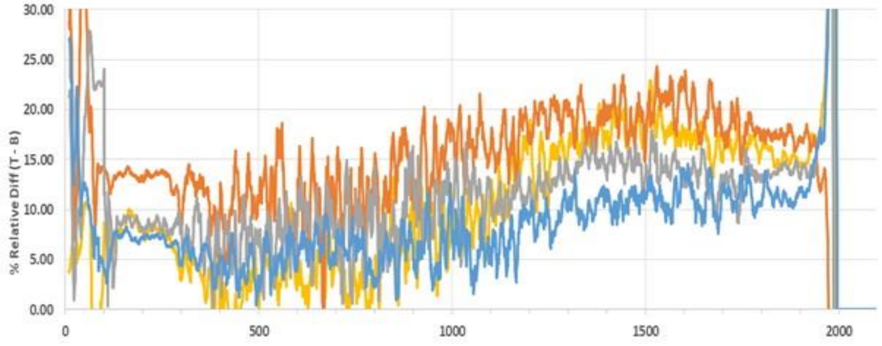
Wind and Weather Conditions: 3/6 wind SSE 2.5 m/s Cloudy and foggy conditions, cleared up by noon. 3/10 conditions wind E, some clouds but mostly sunny.

Relative Differences varied from 2-20% on 1st day, 5-15% on the 2nd day, and 5-15% on the 3rd day

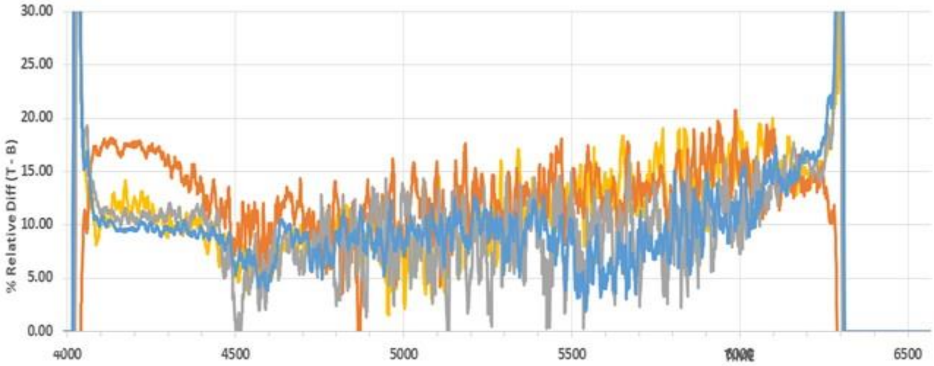
No obvious indications of work-related disturbances, but North and South sides had consistently greatest attenuation.



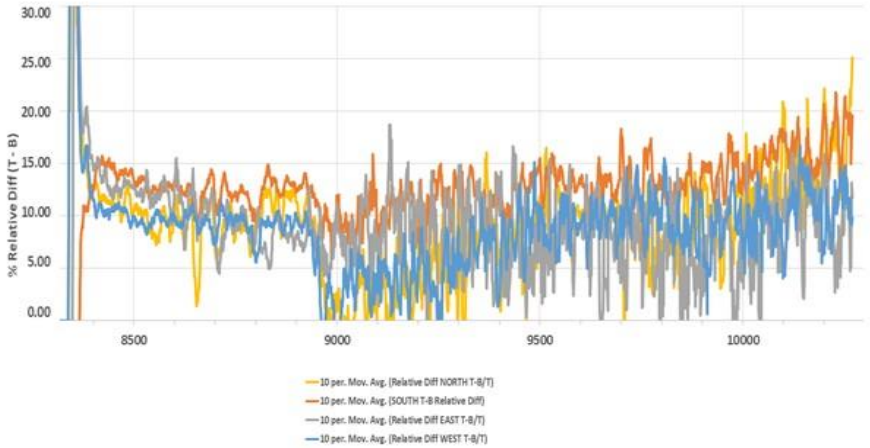
% Relative Difference (Top – Bottom) on 3 Days (Lomax 3/6 – 3/8)



3-6



3-7



3-8

Total Suspended Solids (TSS)

TSS samples were collected upstream and downstream immediately following sediment disturbances. However, the timing of collection varies and may impact the results depending on the speed of the current. Care must be taken to avoid additional sediment disturbances while moving downstream to the collection site. This sort of interference has been detected on several occasions.

Because every experiment was unique, our emphasis here was to look for patterns or trends, and to report the range of numerical values and the means or medians. Generally, it was expected that suspended sediments would be greatest nearest the disturbance and lower with distance away. But delayed collections can result in lower levels near the disturbance and greater levels down current. Two metrics from TSS used for analysis in this report are sediment abundance (dry weight) in mg/L, and % Organic Matter (% OM) of the captured solids.

Note that in these figures, "T" numbers refer to test disturbances and letters A-C refer to lines of measuring stations (Tracks)

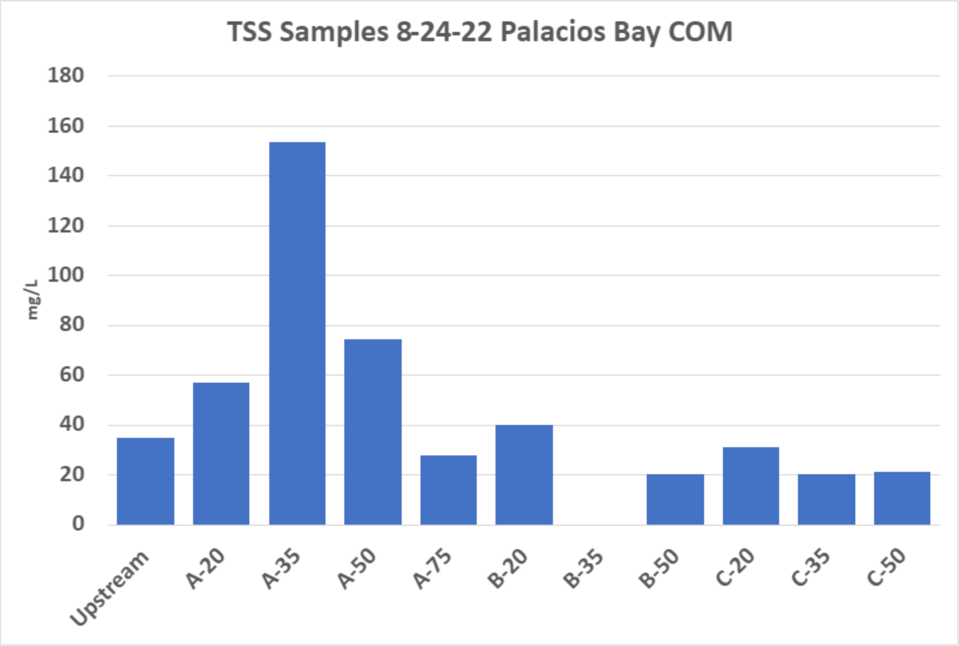
TSS (mg/L) from Shallow COM Sites

Palacios HRI COM 8/24/2022

The expected pattern outlined above appears to be the case for Track A at Palacios HRI COM site on 8-24, and to a lesser extent Tracks B and C. The light loggers for this sampling indicated that Track A was most weakly affected by the sediments compared to the other Tracks. That would be contradictory to these TSS results unless there is a contamination issue. Sediment traps for this sampling (shown below) exhibit the expected relationship with distance, so we conclude that these TSS samples were contaminated.

Conditions: Windy, Cloudy, ~2.1 m/s ~NE wind

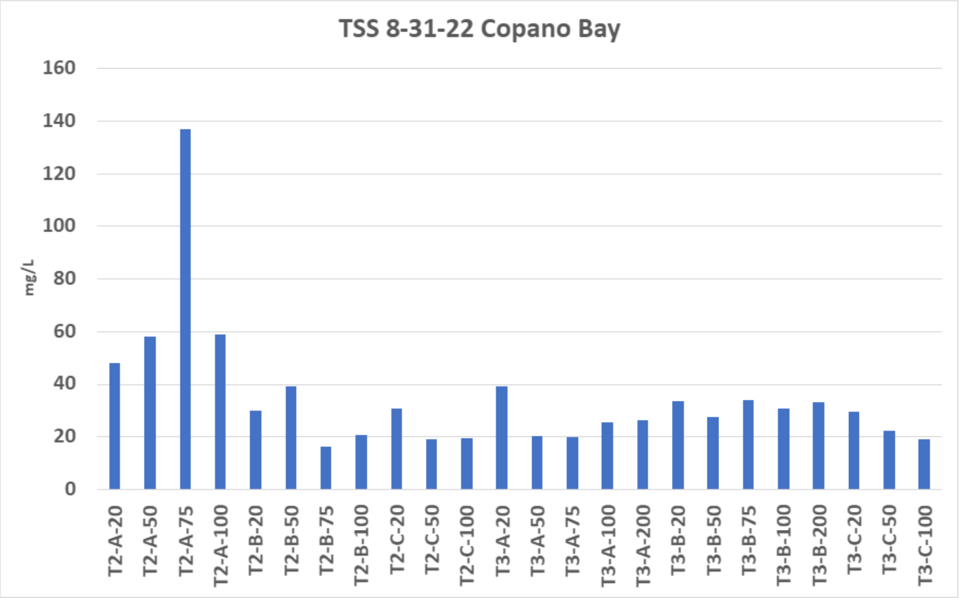
Peak TSS observed at 35 ft downstream, with expected decreasing pattern beyond that distance.



8/31/2022 - Copano Bay HRI COM

Three sediment disturbances were performed at Copano HRI COM site on 8/31/22. The expected relationship with distance was observed only in the case of Track C for both Tests 2 and 3. Tracks A and B deviated somewhat. Also, TSS levels here at this sandy site were generally lower than those observed at other sites.

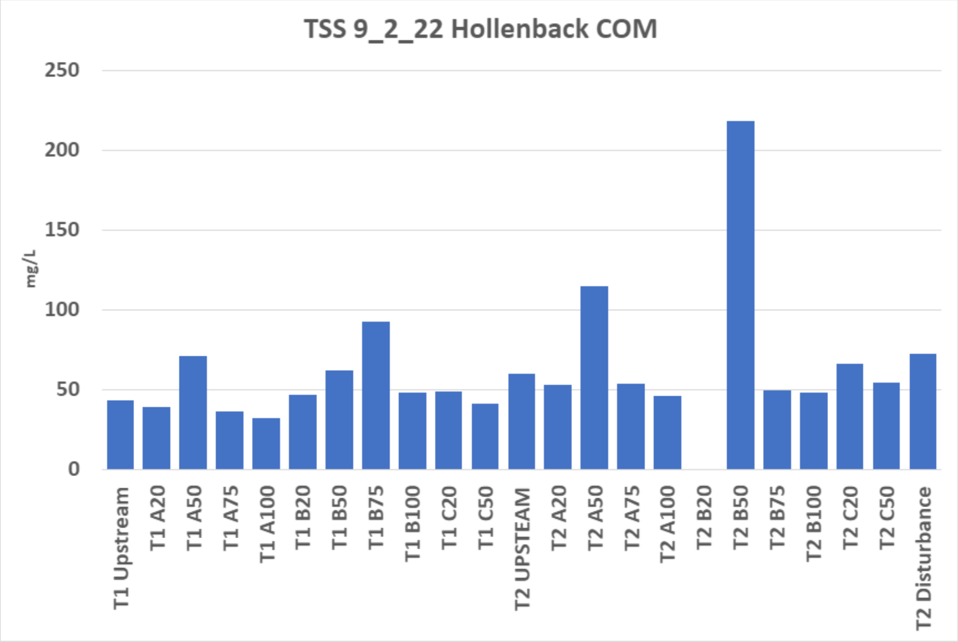
Conditions: Cloudy, Light winds; ~10-12mph E wind; 270 degrees current (westerly)



9/2/2022 - Hollenbeck Site

The 9-2 sampling at the Hollenbeck COM consisted of a series of 3 simulated sediment disturbances, designated T1 – T3 (Test numbers) that were of 45 p-m, 90 p-m and 90 p-m, respectively. Measuring stations were in 3 Tracks (A-C) at indicated distances (ft) from the disturbance near the cage line. There was significant ambient turbidity, but the downstream TSS levels were greater than the upstream and exhibited the expected declining relationship with distance from the disturbance for Test 1. However, Test 2 levels were higher upstream than downstream at Track A, but downstream Track B had much higher levels. This probably related to a wind/current shift that increased ambient turbidity and altered the sediment plume trajectory.

Conditions: Mostly clear; ~4 m/s ESE wind; ESE current; Later in the day went down to ~3m/s

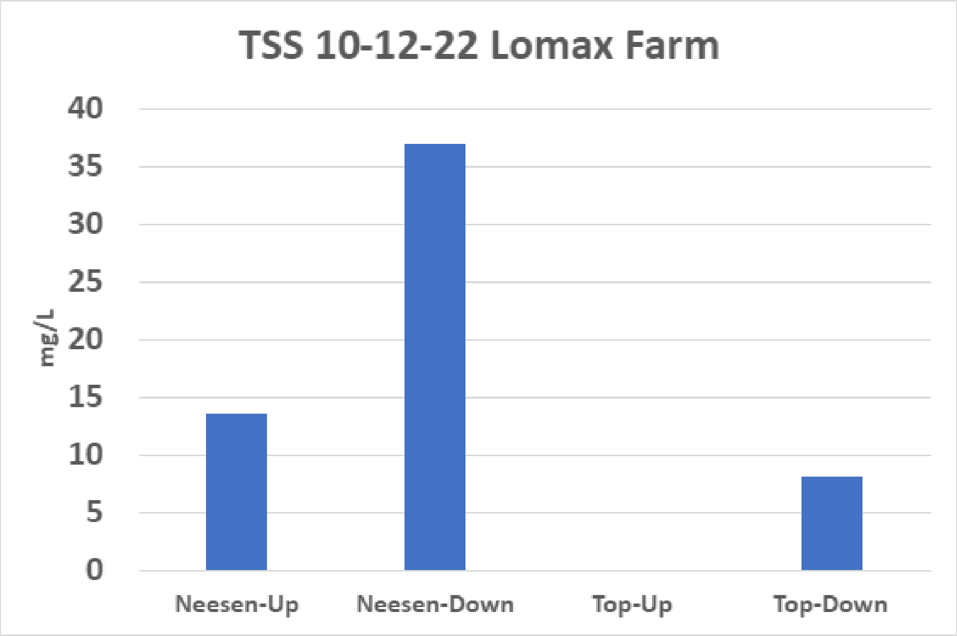


TSS from Lomax COM Multiple Dates

TSS levels observed at the deep water COM site were much lower than observed for the shallow water sites. The currently operating deep water COMs are located in approximately 8 ft of water, so all operation and maintenance tasks are performed from boats, which avoids or minimizes disturbance of the sediments below. The only observable sediments produced in the operations have been the release of the epibiota that accumulates on the oyster cage gear when it is handled for cage flipping, bag shuffles, etc. These actions knock off some of the algae and, perhaps just as importantly, any suspended particulate matter from the water column that became entrained in these slimy biofilms. Observed levels of TSS ranged from 5 to 35 mg/L, substantially below many of those observed at shallow water COM sites.

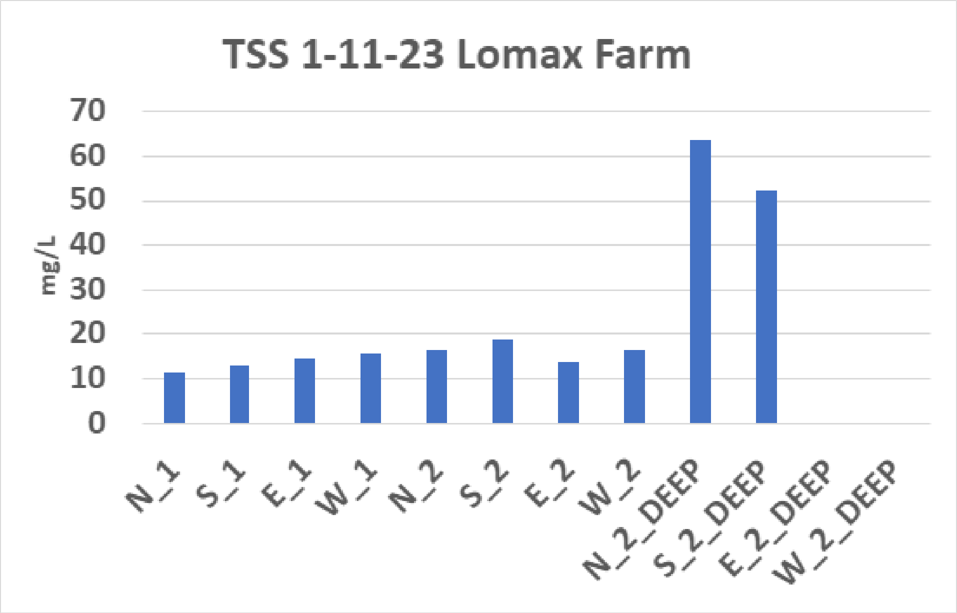
10-12-22 Conditions: Mostly Sunny, Calm; ~<5m/s S wind ; Later ~4m/s East Wind.

For bottom samples (Neesen) the down-current (North) had the greatest level of TSS. The up-current surface water sample was lost in processing.



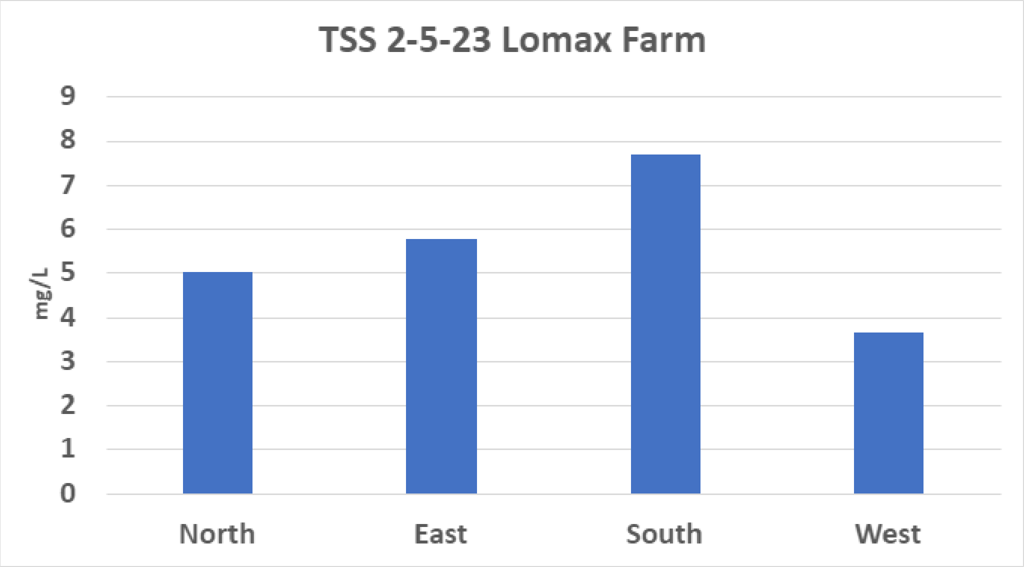
1-11-23 Conditions: ~6.2 m/s S rising to 6.5 m/s S to SSW wind at 210°

South site would be up-wind and up-current and, to lesser extent also the “West”. North would be the main down-current site, and East to a lesser extent. The south and East station poles had some mud on them so they were on or near the bottom. Samples labeled “Deep” were acquired with Neesen sampler on bottom. All other TSS samples from surface water. Note upwind sites had greatest TSS levels, but down-current bottom sample was highest (North).



TSS collections increased later in day (2nd sampling) as the wind increased. The South collection site had the highest TSS level, except for bottom water samples collected with the Neesen sampler.

2-5-23 Conditions: Flat Water, Calm and Clear, 5mph wind out of the Northeast; Then shifting to SE and rising at midday to about 9 mph. If most TSS came from the stronger wind period in afternoon, then South and East samples would be up-current.



2-21-23 to 2-24-23 Multiday Sampling at Deep Water Lomax COM Site

Conditions: 2-21: Cloudy and Foggy, 14mph Wind SSE; Later ~5.5 m/s S Wind (190 degrees),

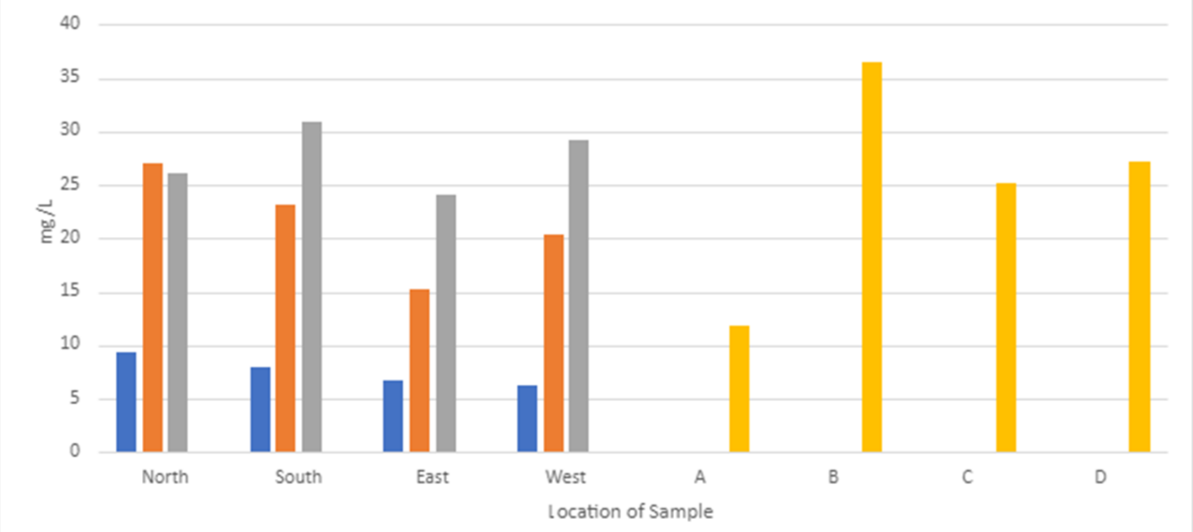
Conditions 2-22, Wind out of southwest ~ 14 mph shifting to shifting to southeast ~10 mph early afternoon, Employees worked 10:00 AM – 2:00 PM

Conditions 2-23, Winds out of South-Southeast ~ 8 mph, shifting to East 10-12 mph at midday, Employees worked 9:30 AM – 1:45 PM

Conditions: 2-24: ~4.8 m/s out of the E to ESE (115 degrees). Employees worked 9:30 AM – 2:00 PM

Blue bars – 2/21; Orange bars – 2/24 Pre-work samples in morning; Gray bars – 2/24 Post-work samples in afternoon. Yellow bars are supplemental TSS collections from near or over the cages when they were flipped back into the water. These values would presumably represent the greatest concentration of TSS possibly released from a cage-flip. For 2-24-23, solids collected at-cages during COM procedures (yellow) were on average only slightly higher than background (Pre, orange) TSS levels but less than the average of values collected at sampling stations post, and thus little different from ambient water TSS around the farm.

Mean of 4 samples collected Pre-, during, and Post- were, respectively, 21.9, 25.4, and 27.8 mg/L (no significant difference).



Variations between 2/21 and 2/24 samples could be result of varying weather conditions on the two days, but relationships between the collecting sites were mostly similar, with North and South sites highest.

3/6/23 to 3/9/23 Multiday Sampling at Deep Water Lomax COM Site

Conditions 3/6: wind SSE 2.7 m/s (150°)Cloudy and foggy conditions, cleared up by noon, winds increased to ~12 mph. Instruments deployed in morning. Workers harvested ~1000 oysters from research row 7 and tumbled 3 cages (morning & afternoon)

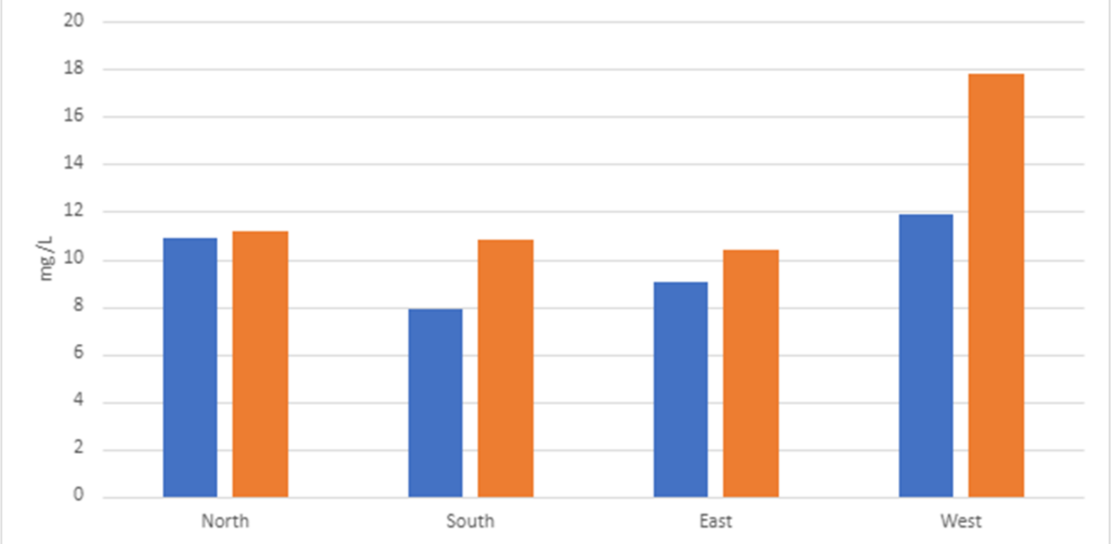
Conditions 3/7: wind SSE to SE increased from ~ 6 mph in morning to ~15 mph in afternoon. Workers tumbled 2 research cages and dessicated 3 cages in row 4.

Conditions 3/8: winds ~ 10 mph SE increased to ~ 14 mph SE in afternoon. Workers harvested ~1500 oysters and re-flipped 3 cages from row 4, then tumbled two cages and returned to row 4.

Conditions 3/9: winds SSE to SE 10-14 mph. Tumbled 1 cage in morning and toured visitors around noon – weather rough.

Conditions 3/10: winds 2.5 m/s at 60°-65° ENE in morning. All measuring stations pulled by 9 AM in morning and TSS samples collected.

Blue bars – 3/6; Orange bars – 3/10 TSS samples in mornings;



The western station (down-current on 3/10; orange) had higher levels of TSS on 3/10 than other stations on the same day. For 3/6 (blue) North and West stations were slightly higher and down-current on that day. Overall there was low variation between stations on the two sample collection dates.

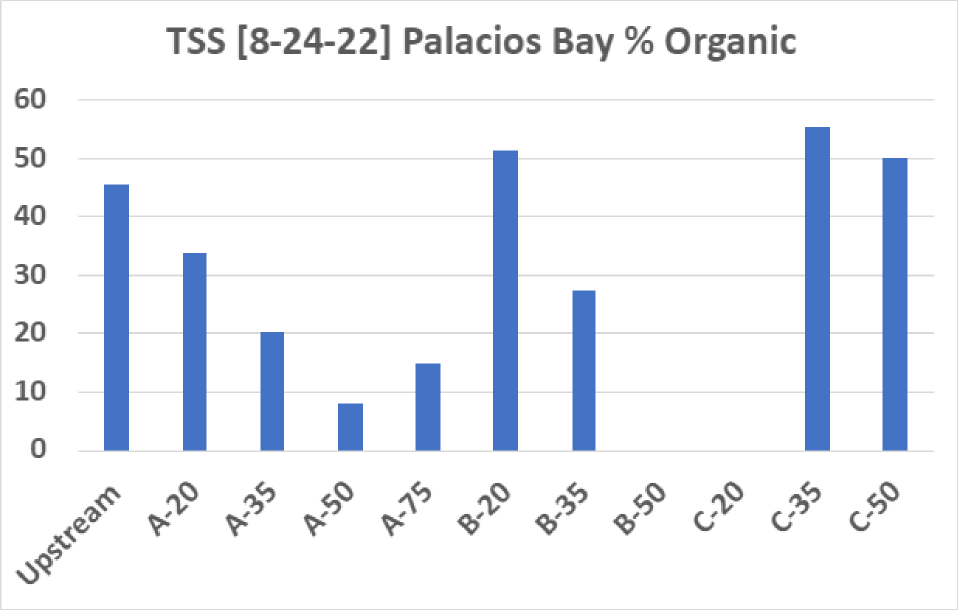
% Organic Matter in TSS

Dried TSS and sediment trap samples were routinely subjected to ashing to remove the organic carbon. This results in a weight change proportional to the organic C content of the sample, and the % of the dry weight lost upon ashing provides insight into the relative proportions of organic matter (OM) vs inorganic content. It is expected that sandy sediments will exhibit low % OM, whereas material derived from non-calcareous biota will have a relatively high % OM. Also, it is predicted that suspended sandy sediments will settle quickly, or near the disturbance, while organic particles are likely to stay suspended longer and travel farther. So % OM provides an important characterization of the suspended and settled solids.

TSS % OM from Shallow COM Sites

Palacios HRI COM 8/24/2022

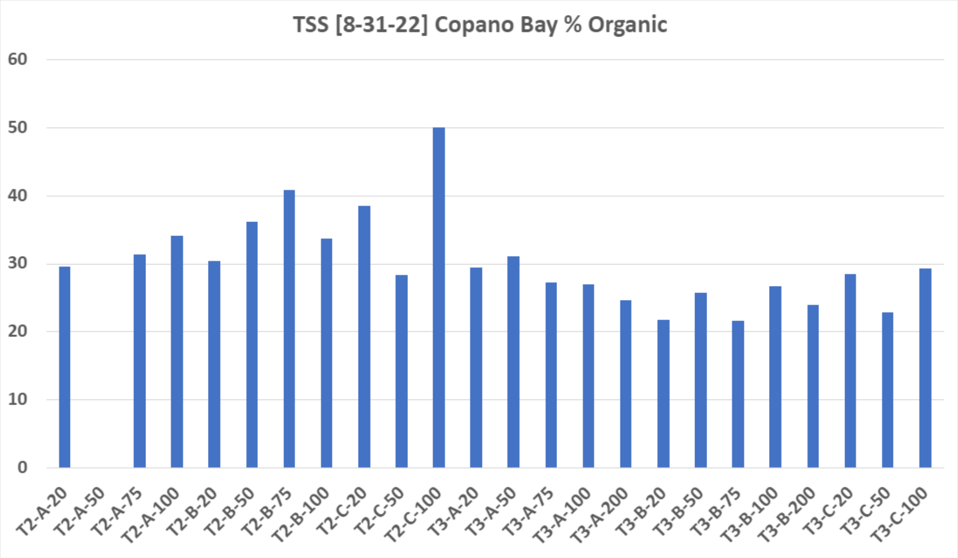
Conditions: Windy, Cloudy, ~2.1 m/s all day but direction shifted from S early to NE for expts



Upstream samples had high %OM levels, which lowered on the down current side of a disturbance and further diminished with distance. This was inconsistent with expectations.

8/31/2022 - Copano Bay HRI COM

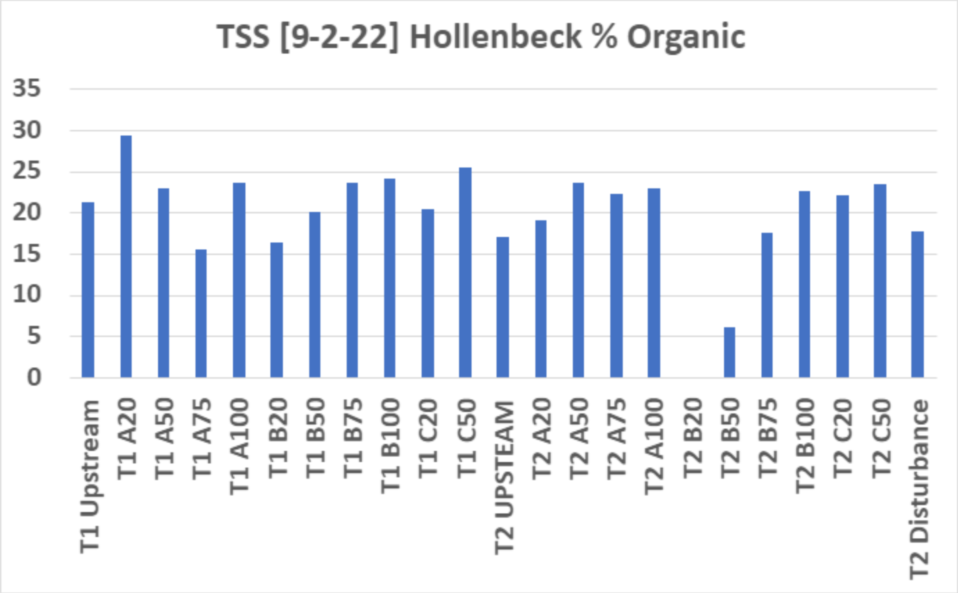
Conditions: Cloudy, Light winds; ~10-12mph E wind; 270 degrees current



The percent organic content of the TSS was mostly between 20-40%, but no clear patterns were evident.

9/2/2022 - Hollenbeck Site

Conditions: Mostly clear; ~4 m/s ESE wind; ESE current; Later in the day went down to ~3m/s

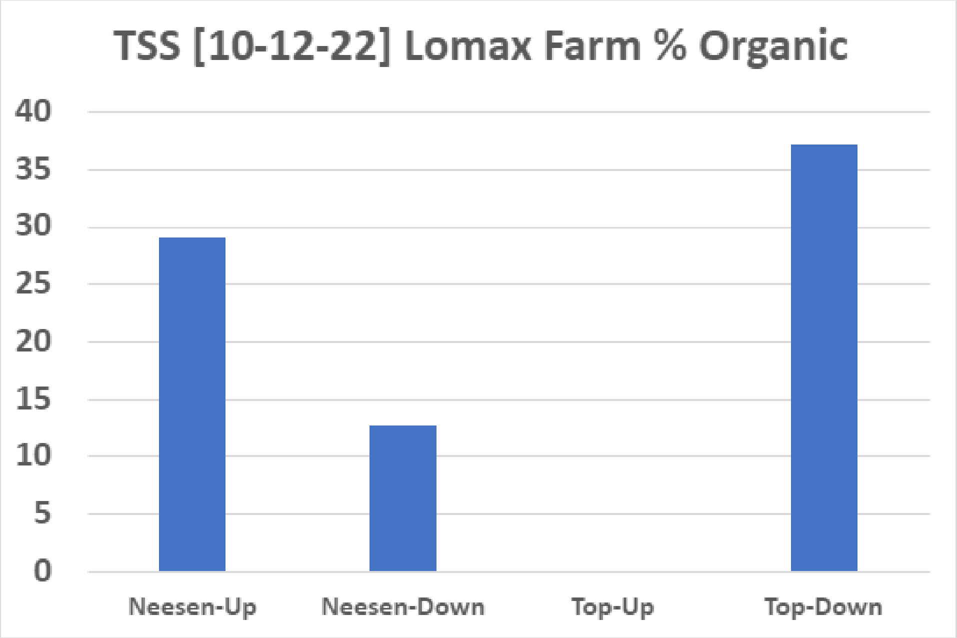


For this experiment % OM varied from 15% - 30% and, in most cases, there was an increase in %OM with distance down-current, consistent with expectations.

TSS % OM from Lomax COM Multiple Dates

10-12-22 Conditions: Mostly Sunny, Calm; ~<5m/s S wind ; Later ~4m/s East Wind.

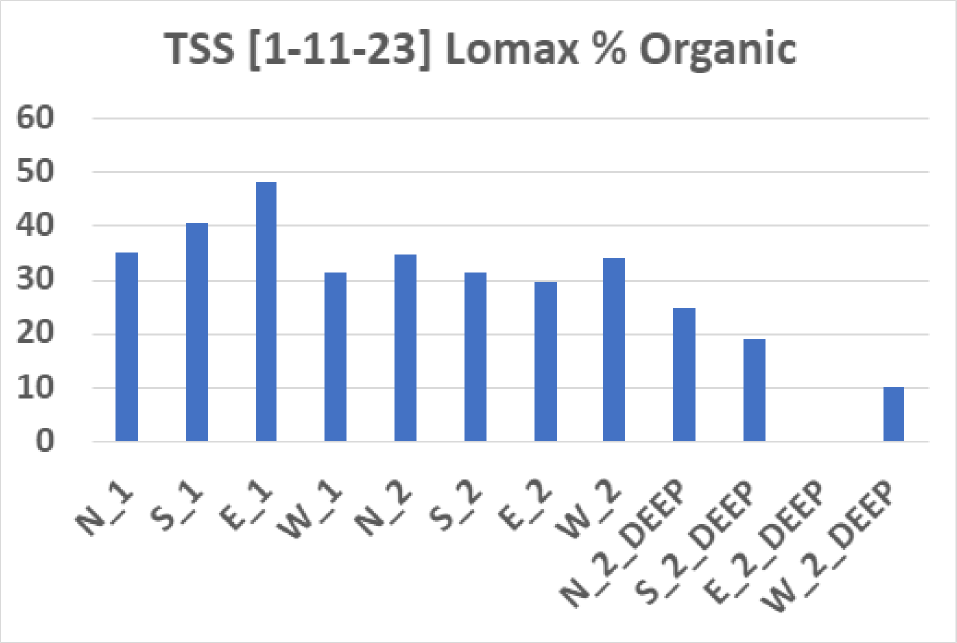
For bottom samples (Neesen) the down-current (North) had the greatest level of TSS. The up-current surface water sample was lost in processing.



The pattern here is opposite to that observed for TSS values. The down-current (North) had the greatest level of TSS but the lowest % OM here. The higher % OM values were obtained for the lowest TSS collections.

1-11-23 Conditions: ~6.2 m/s S rising to 6.5 m/s S to SSW wind at 210°

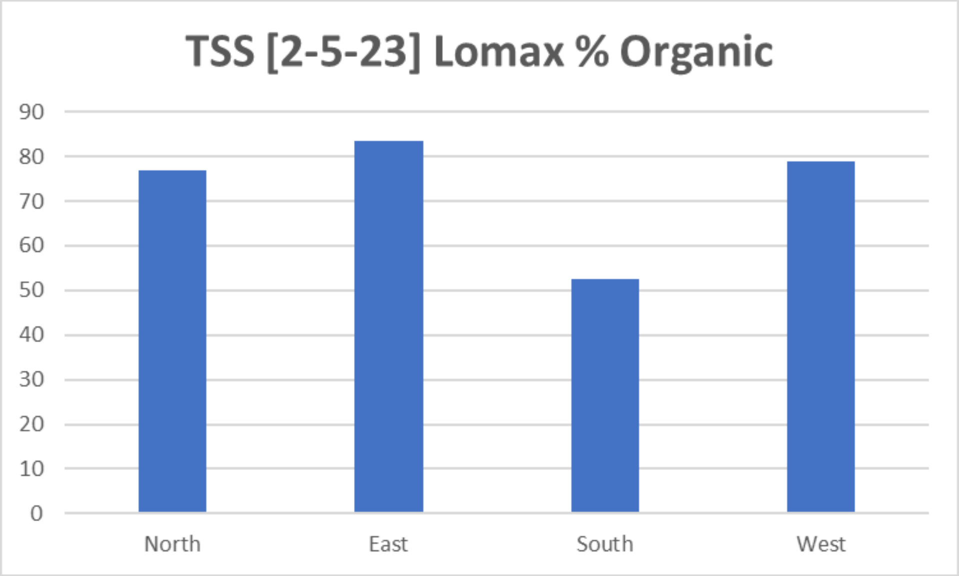
South site would be up-wind and up-current and, to lesser extent also the “West”. North would be the main down-current site, and East to a lesser extent. The south and East station poles had some mud on them so they were on or near the bottom. Samples labeled “Deep” were acquired with Neesen sampler on bottom. All other TSS samples from surface water. Note upwind sites had greatest TSS levels, but down-current bottom sample was highest (North).



The percent OM in surface TSS samples ranged from 20-50%, and lower values observed for deep samples near the bottom. No directional trends evident.

2-5-23 Conditions: Flat Water, Calm and Clear, 5mph wind out of the Northeast; Then shifting to SE and rising at midday to about 9 mph. If most TSS came from the stronger wind period in afternoon, then South and East samples would be up-current.

Unusually high % OM values (50 - 85%). The up-current South sample had lowest % OM.



2-21-23 to 2-24-23 Multiday Sampling at Deep Water Lomax COM Site

Conditions: 2-21: Cloudy and Foggy, 14mph Wind SSE; Later ~5.5 m/s S Wind (190 degrees),

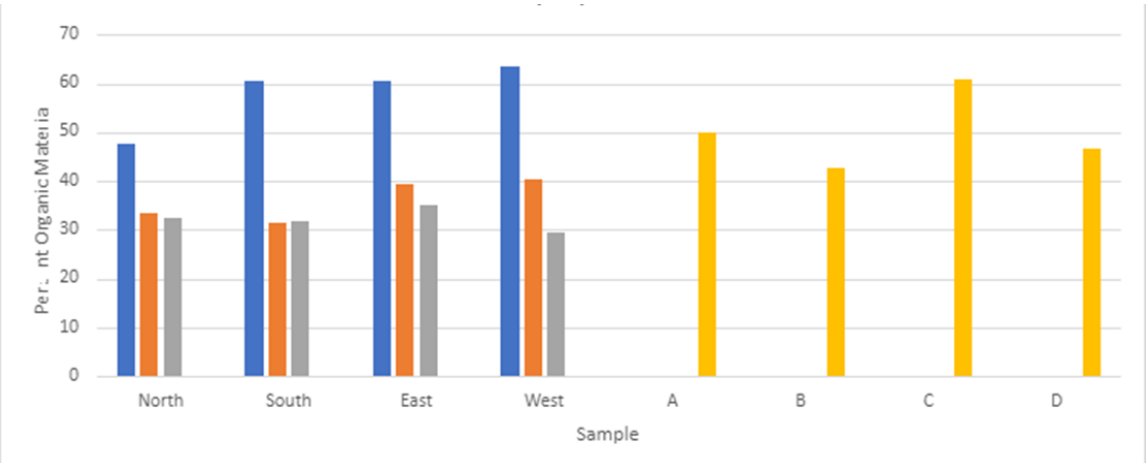
Conditions 2-22, Wind out of southwest ~ 14 mph shifting to shifting to southeast ~10 mph early afternoon, Employees worked 10:00 AM – 2:00 PM

Conditions 2-23, Winds out of South-Southeast ~ 8 mph, shifting to East 10-12 mph at midday, Employees worked 9:30 AM – 1:45 PM

Conditions: 2-24: ~4.8 m/s out of the E to ESE (115 degrees). Employees worked 9:30 AM – 2:00 PM

Percent Organic Matter in 2/21 and 2/24 TSS Samples.

Blue bars – 2/21; Orange bars – 2/24 Pre-work samples in morning; Gray bars – 2/24 Post-work samples in afternoon. Yellow bars are supplemental TSS collections from near or over the cages when they were flipped back into the water. These values would presumably represent the greatest concentration of TSS possibly released from a cage-flip, and we expect these samples to contain the greatest levels of % OM as they should be mostly comprised of epibiota.



The near-cage samples had a higher % OM compared to TSS samples from measuring stations on the same day. But these values were still less than observed for station TSS samples on 2/21.

The average % OM values for 2/21 Pre (blue), 2/24 Pre (orange), 2/24 Post (gray), and 2/24 near-cage samples (yellow) were 57.9%, 36%, 32%, and 50.5%, respectively.

Again, see pattern where lowest TSS collections had the greatest % OM, within the range of 30% – 65% OM.

Settled Sediments (Sediment Traps)

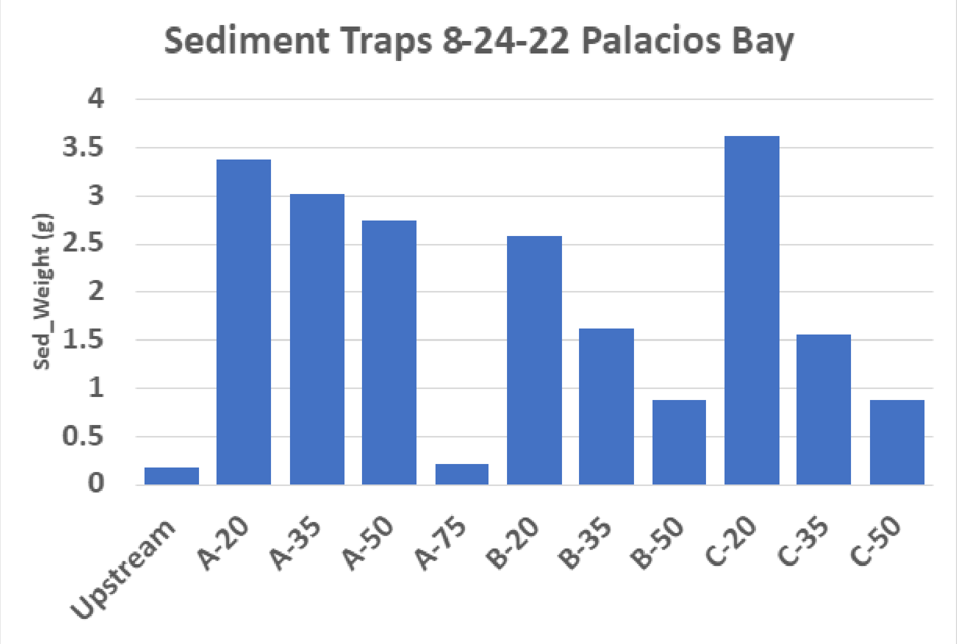
Settled Sediment mass as g per daily deployment period (typically 3-4 hr)

Sediment Traps from Shallow COM Sites

Sediment traps capture the readily settleable suspended solids. A gradient of decreasing sediment collection is expected with distance from the disturbance.

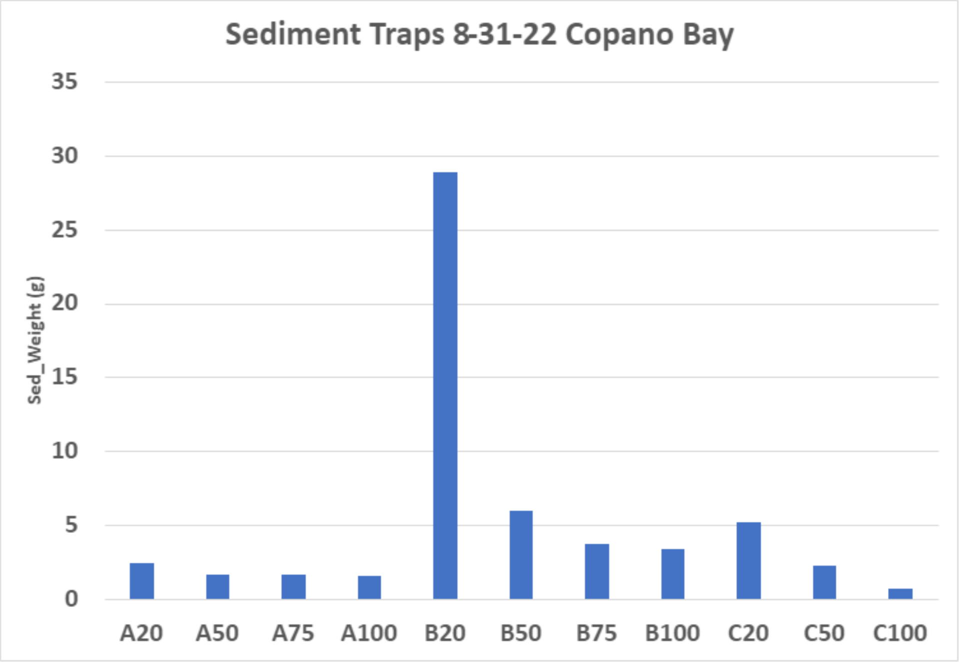
All 3 tracks at Palacios HRI COM and Copano HRI COM showed this relationship. In contrast, the 9-2-22 experiment at Hollenbeck COM yielded inconsistent results. Sediment trap collections mostly less than 5 g dry weight for 3 disturbances in one day.

8/24/22 Conditions: Windy, Cloudy, ~2.1 m/s NE for expts



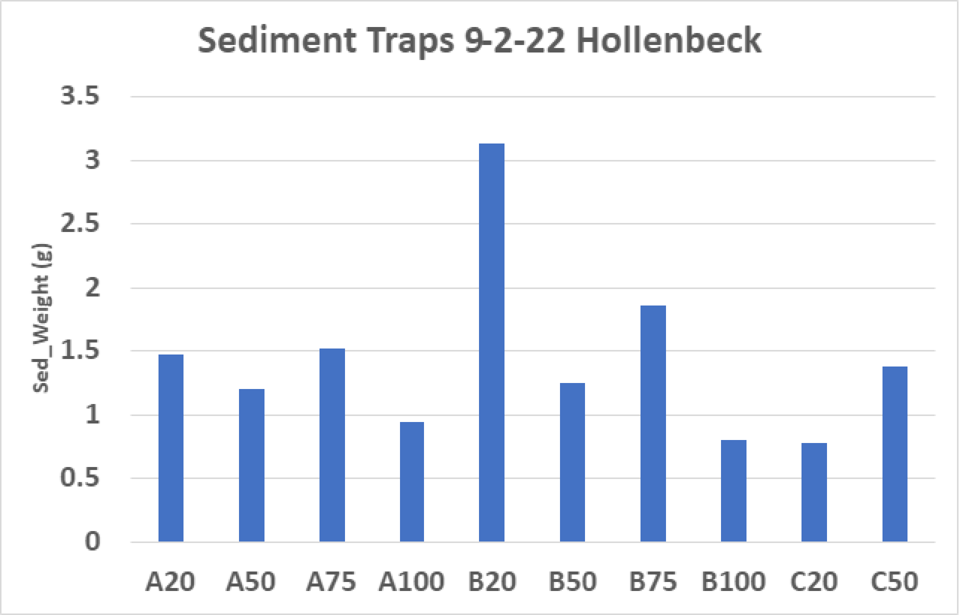
8/24/22 sampling at Palacios Bay resulted in a clear gradient of sediment deposition across distance (as espected). With A, B, and C tracks receiving a higher amount of sediment in the traps nearest to the cages and decreasing on a gradient as distance from the cage increased. All 3 tracks were placed down current of the oyster cages offset by ~30 degrees to account for any variations or changes in current. Low sediment trap collections were observed upstream and at the farthest location downstream (Track A 75 ft).

8-31-22 Conditions: Cloudy, Light winds; ~10-12mph E wind; 270 degrees current



Observe very small but consistent downward trend of collected sediments with distance. Track B better aligned with sediment plumes than Track A. Not clear if B 20 ft collection is an artifact or representative of being very near to the disturbance.

9-2-22 Conditions: Mostly clear; ~4 m/s ESE wind; ESE current; Later in the day went down to ~3m/s. Less consistent with expected trend, but similar collection range and a high B 20 value.

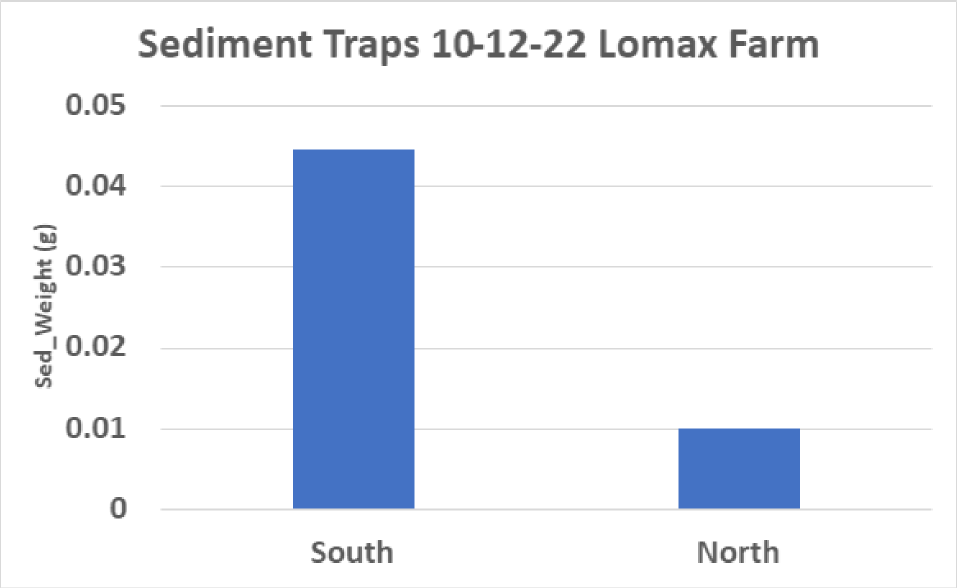


Sediment Traps from Deep COM Sites

For sediment trap collection at the deep water COM site, the collection was performed mid to low in the water column, but not on the bottom as for deployments at the shallow water COM sites.

There seems to be one side that collects a large amount, and the other 3 sides collect very little. Note that all sediment trap collections at this deep COM site were much lower (< 1 g) than for the simulated disturbances at the shallow COM sites (typically 1 – 5 g) for approximately similar periods of time.

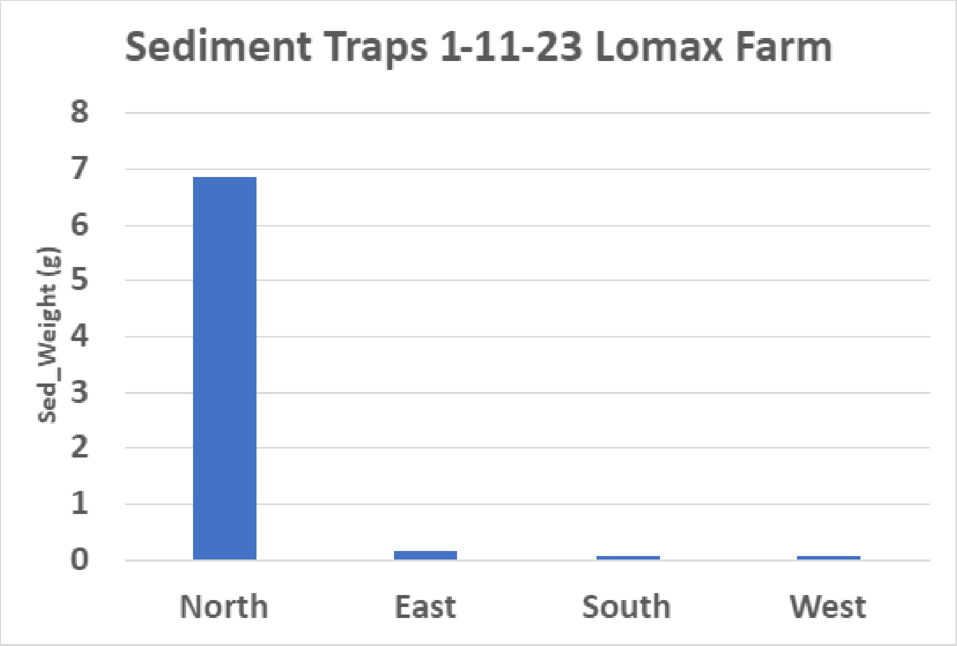
10-12-22 Conditions: Mostly Sunny, Calm; ~<5mph S wind ; Later ~4m/s East Wind.



The South collection was the up-current side this day, and had a greater collection level.

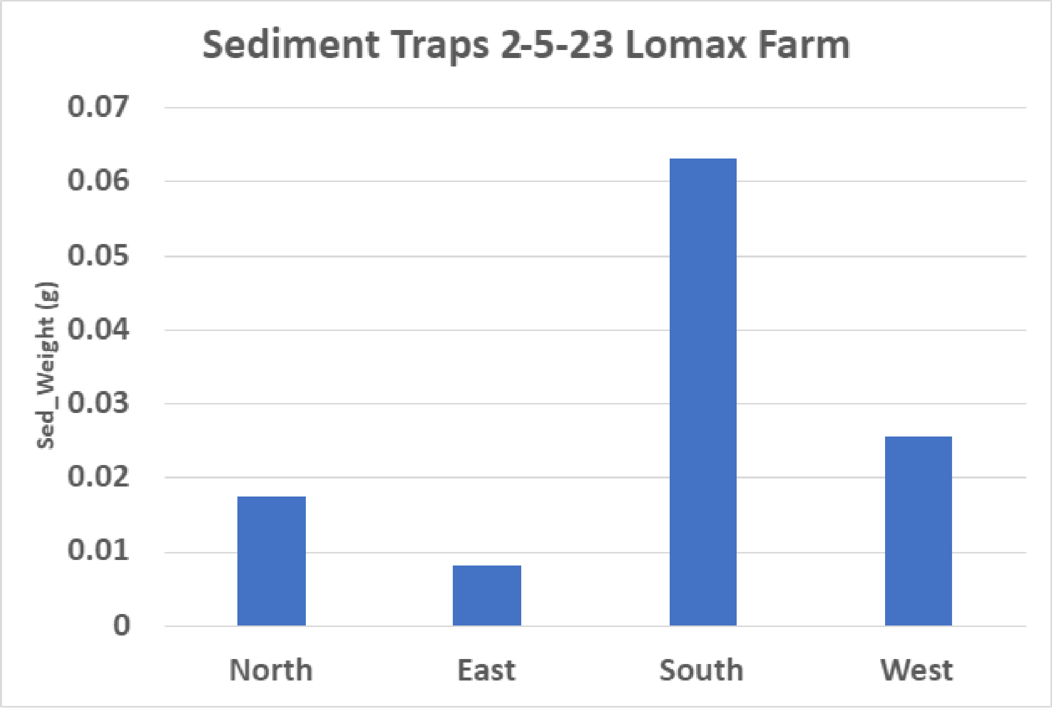
1-11-23 Conditions: ~6.2 m/s S rising to 6.5 m/s S to SSW wind at 210°

South site would be up-wind and up-current and, to lesser extent also the “West”. North would be the main down-current site, and East to a lesser extent. The south and East station poles had some mud on them so they were on or near the bottom.



Not clear why North collection is at least 10-fold higher than all others at this COM site, but this was the down-current side. Possibly artefact from dipping into mud bottom.

2-5-23 Conditions: Flat Water, Calm and Clear, 5mph wind out of the Northeast; Then shifting to SE and rising at midday to about 9 mph. If most settleable solids came from the stronger wind period in afternoon, then South and East samples would be up-current.



South side (up-current) had the greatest level here, but total collection values very low on this relatively calm day.

2-21-23 to 2-24-23 Multiday Sampling at Deep Water Lomax COM Site

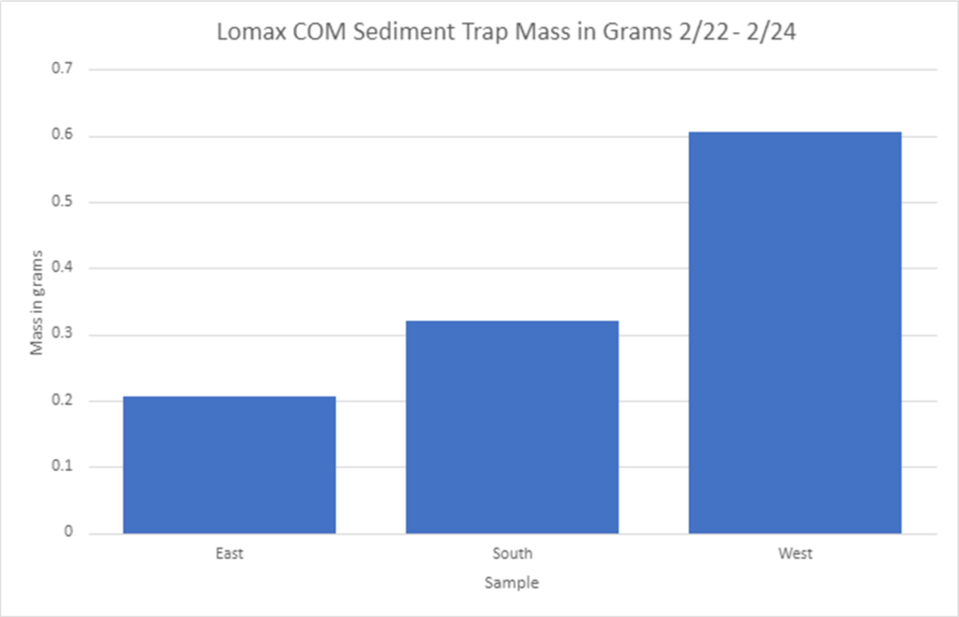
Conditions: 2-21: Cloudy and Foggy, 14mph Wind SSE; Later ~5.5 m/s S Wind (190 degrees),

Conditions 2-22, Wind out of southwest ~ 14 mph shifting to shifting to southeast ~10 mph early afternoon, Employees worked 10:00 AM – 2:00 PM

Conditions 2-23, Winds out of South-Southeast ~ 8 mph, shifting to East 10-12 mph at midday, Employees worked 9:30 AM – 1:45 PM

Conditions: 2-24: ~4.8 m/s out of the E to ESE (115 degrees). Employees worked 9:30 AM – 2:00 PM

The sediment traps were deployed for 4 days here, so for comparisons to other samples, the sediment dry weights should be divided by 4 to get a daily collection value. When this is done, the collected values range from 0.05 to 0.12 g per trap (0.0088 m^2) per day, which are similar to other collection values. The weather varied widely, from calm to strong winds, and the North trap was lost over the sampling period.



The greatest level of observed sediment deposition was at the western sediment trap, more on the down-current side, along with North site (for which the data was not collected), of the tracks for this sampling period.

Conditions 3/6: wind SSE 2.7 m/s (150°) Cloudy and foggy conditions, cleared up by noon, winds increased to ~12 mph. Instruments deployed in morning. Workers harvested ~1000 oysters from research row 7 and tumbled 3 cages (morning & afternoon)

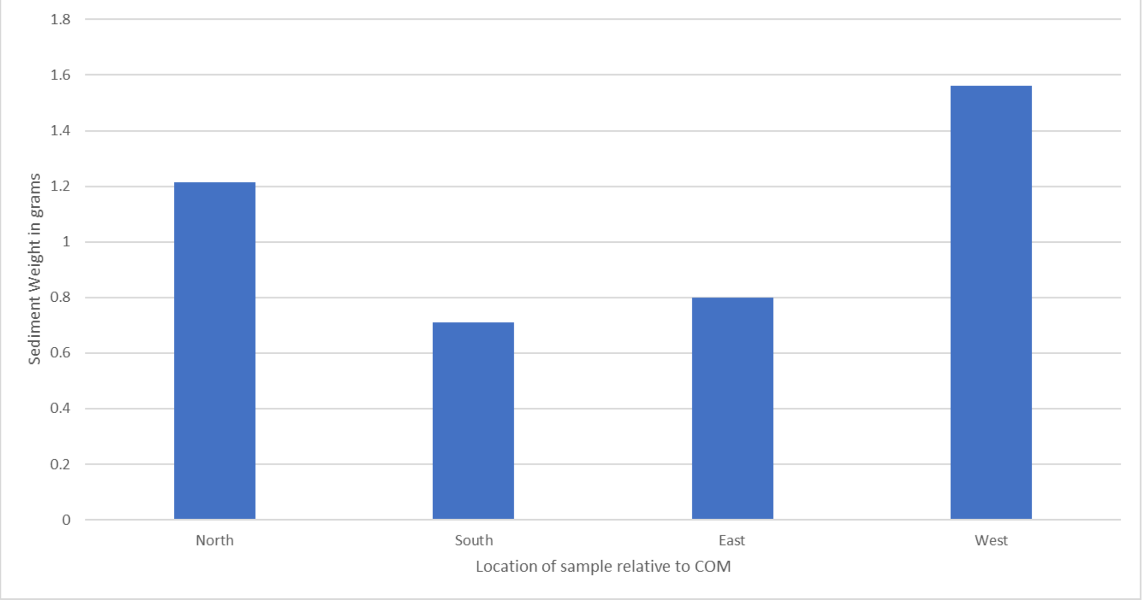
Conditions 3/7: wind SSE to SE increased from ~ 6 mph in morning to ~15 mph in afternoon. Workers tumbled 2 research cages and dessicated 3 cages in row 4.

Conditions 3/8: winds ~ 10 mph SE increased to ~ 14 mph SE in afternoon. Workers harvested ~1500 oysters and re-flipped 3 cages from row 4, then tumbled two cages and returned to row 4.

Conditions 3/9: winds SSE to SE 10-14 mph. Tumbled 1 cage in morning and toured visitors around noon – weather rough.

Conditions 3/10: winds 2.5 m/s at 60°-65° ENE in morning. All measuring stations pulled by 9 AM in morning.

Blue bars: 3/10 Post 4 day sample collection of sediment trap settleable solids.



The down-current locations North and West had the greatest sediment collections for the 4-day period. Dividing these values by 4 to get a daily collection value leads to a range of 0.15 to 0.4 g per trap per day.

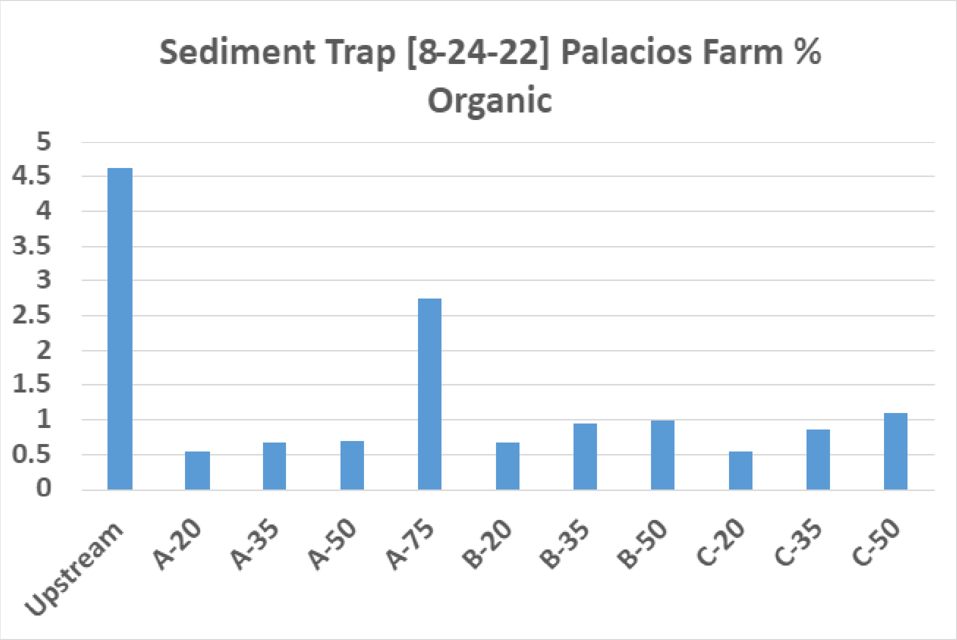
% Organic Component (Sediment Traps)

As for TSS, it is expected that in sandy sediments, settled particulates will exhibit low % OM, whereas material derived from non-calcareous biota will have a relatively high % OM. Also, it is predicted that suspended sandy sediments will settle quickly, or near the disturbance, while organic particles are likely to stay suspended longer and travel farther. This would predict a gradient of increasing % OM with distance from the disturbance. So % OM provides an important characterization of the settled solids.

Shallow COM Sites

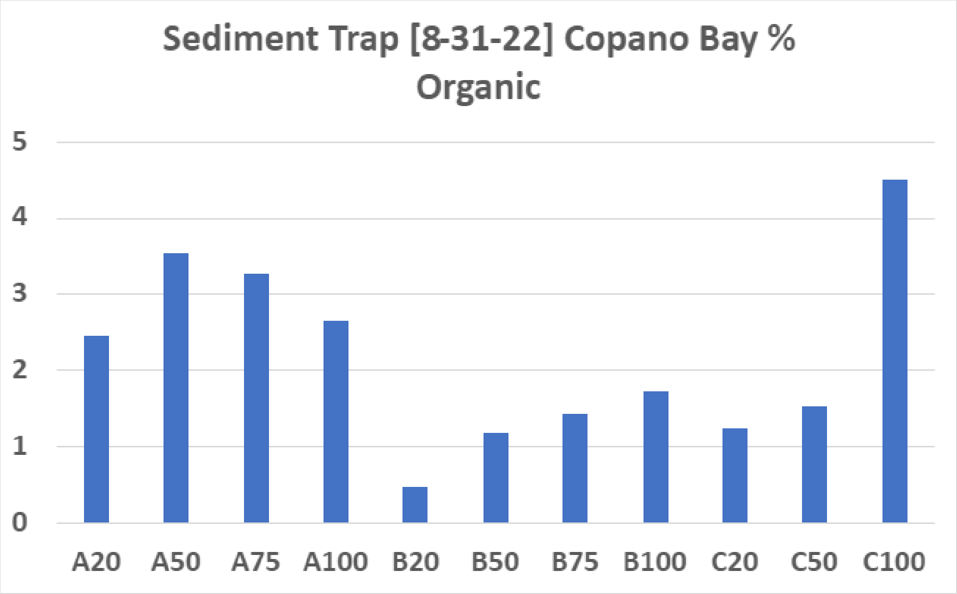
The %OM at Palacios COM exhibited a slightly increasing pattern with distance, as expected, for all 3 tracks. The same was generally true at the other shallow COM sites as well. The % OM in settled sediments is much lower than that in TSS samples, so the sediment traps seem to target the heavier sandy particles.

Conditions: Windy, Cloudy, ~2.1 m/s NE for expts



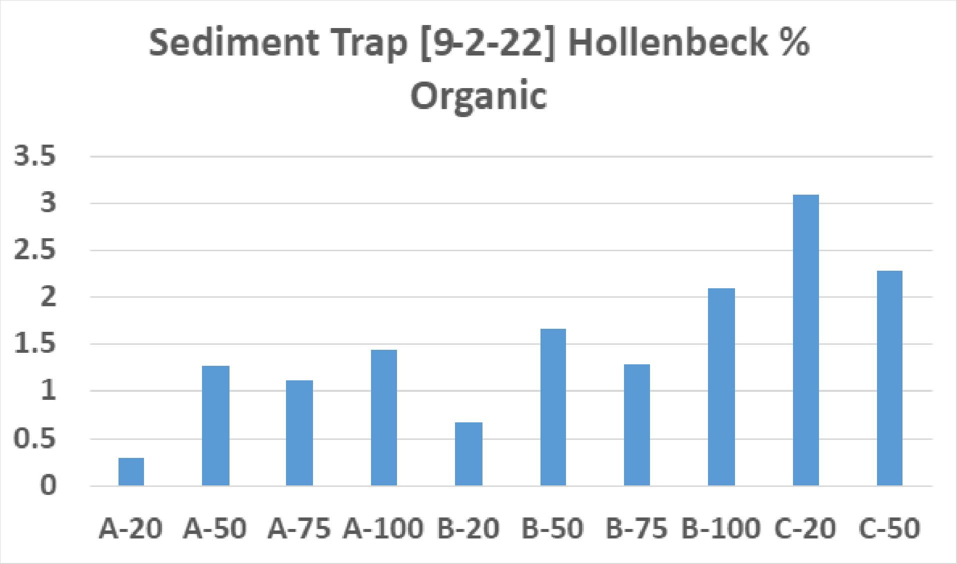
The upstream control sample had the greatest % OM, but this percentage was at least 5-fold lower than observed for TSS sample. The low % organic matter downstream of disturbance indicates most of the settled sediments consisted of sandy or silt bottom sediments, and the increase with distance (albeit very small) for all 3 tracks was consistent with preferential settling of non-organic sediments with distance.

8-31-22 Conditions: Cloudy, Light winds; ~10-12mph E wind; 270 degrees current



Low but generally increasing %OM with distance (Tracks B and C) consistent with preferential settling of non-organic sediments with distance.

9/2/22 Conditions: Mostly clear; ~4 m/s ESE wind; ESE current; Later in the day went down to ~3m/s

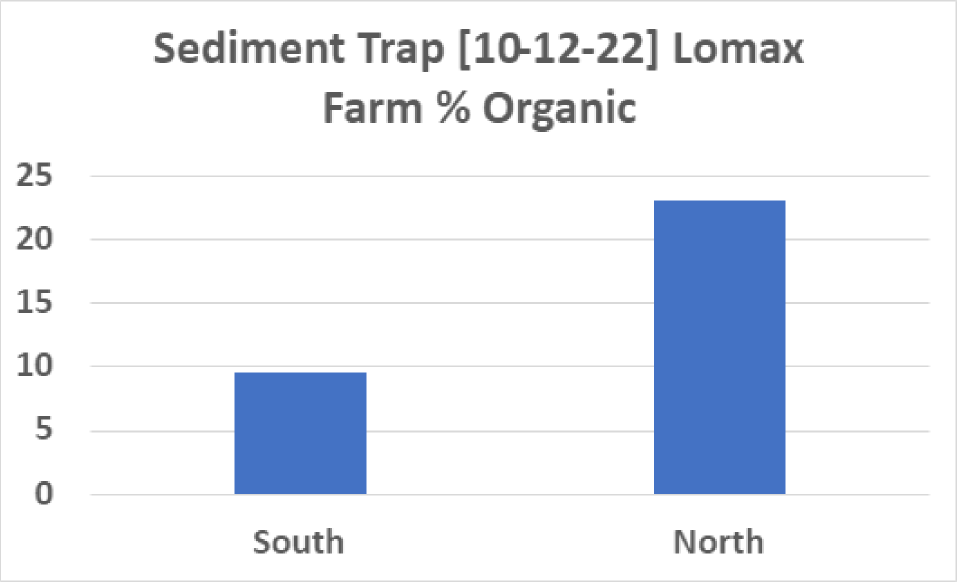


Low but generally increasing %OM with distance consistent with preferential settling of non-organic sediments with distance.

Deep Water COM Site

The % Organic Matter in sediment traps appeared to exhibit an inverse, albeit weak, correlation with sediment weight. Greater sediment weights at the deep water COM site often had a lower percentage of organic material in the sediment trap (Spearman's Rank correlation = -0.134). So where a large amount of sediment is trapped, the %OM is very low. Conversely, for lower amounts of trapped sediments, the %OM was higher. Possible explanations could be larger proportions of water column suspended inorganic matter (sand or calcareous material) becoming entrained in the heavy biofouling communities, or that heavy biofouling communities contain greater proportions of calcareous organisms that are released by cage handling. This is consistent with the sandy nature of particulate matter suspended in the water at Copano Bay. An alternative explanation is that the ashing procedure is not sufficiently thorough for larger samples. We re-ash samples if they look dark after the regular ashing procedure.

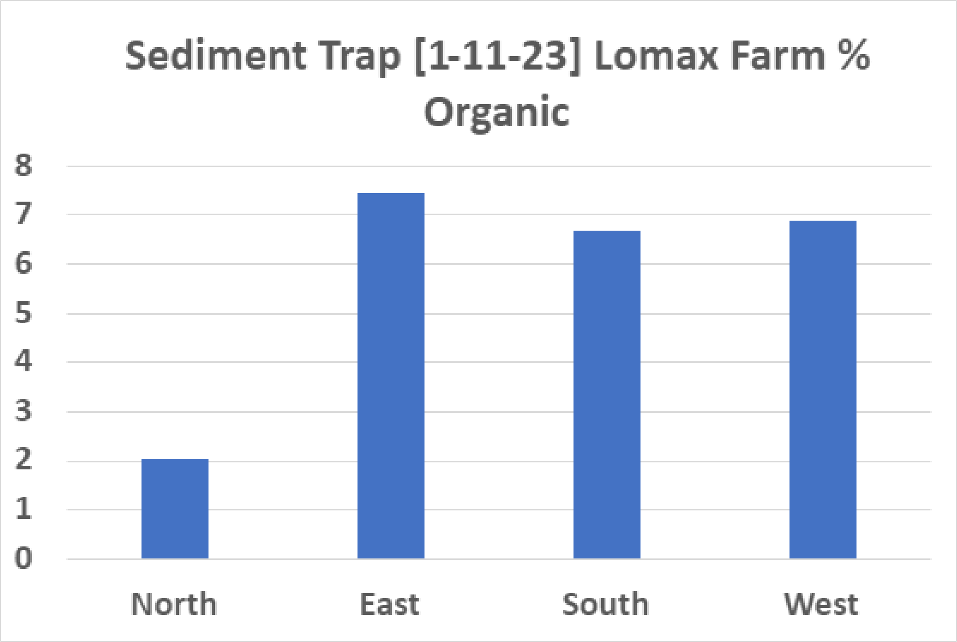
10-12-22 Conditions: Mostly Sunny, Calm; ~<5mph S wind ; Later ~4m/s East Wind.



The North site was partially down-current in the morning and partially up-current for the afternoon winds from the East. North had the higher level of % OM in the settled sediments, but a lower sediment collection compared to South.

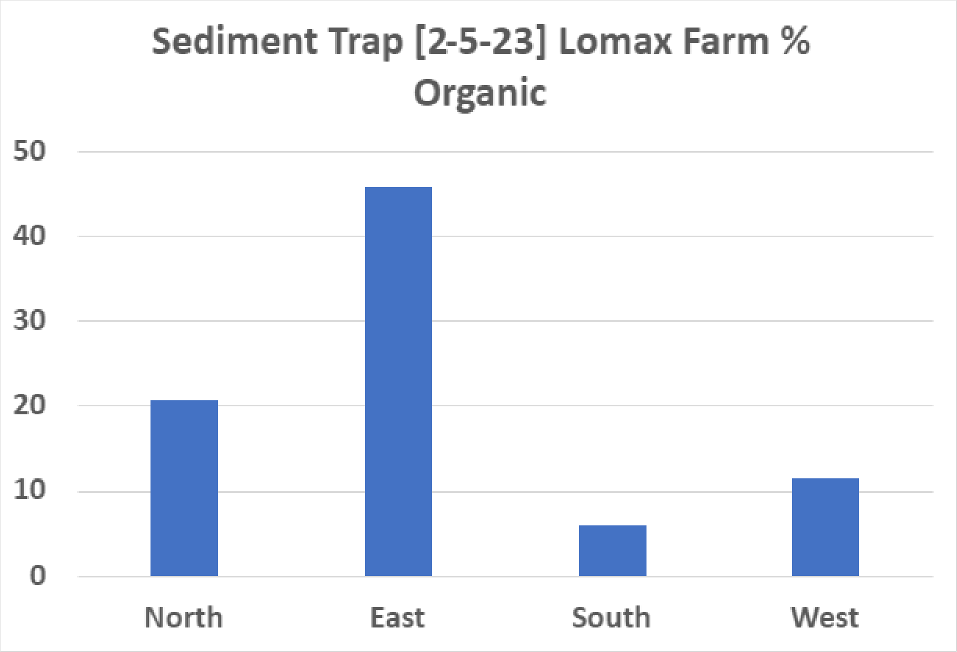
1-11-23 Conditions: ~6.2 m/s S rising to 6.5 m/s S to SSW wind at 210°

South site would be up-wind and up-current and, to lesser extent also the “West”. North would be the main down-current site, and East to a lesser extent.



North and East would be the down-current stations, but North had 3-fold lower % OM along with the largest sediment collection. Sample collection for the North site on this day then had >10-fold increase in sediments (relative to other sites) but the collected material had less than 1/3 the amount of organic material. This would be consistent with contamination by bottom sediments.

2-5-23 Conditions: Flat Water, Calm and Clear, 5mph wind out of the Northeast; Then shifting to SE and rising at midday to about 9 mph.



Again, the lowest % OM (South and West) correlates with the highest sediment collection.

2-21-23 to 2-24-23 Multiday Sampling at Deep Water Lomax COM Site

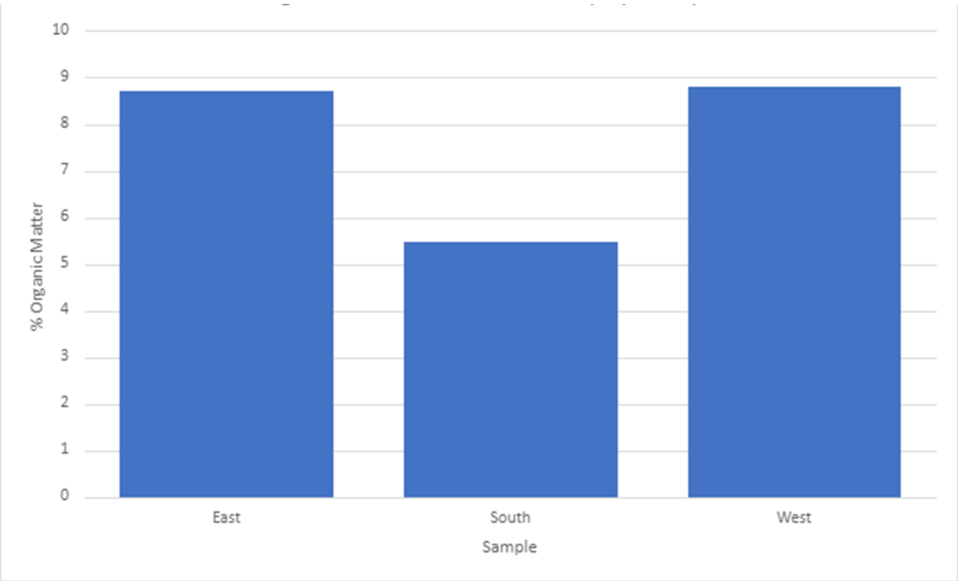
Conditions: 2-21: Cloudy and Foggy, 14mph Wind SSE; Later ~5.5 m/s S Wind (190 degrees),

Conditions 2-22, Wind out of southwest ~ 14 mph shifting to shifting to southeast ~10 mph early afternoon, Employees worked 10:00 AM – 2:00 PM

Conditions 2-23, Winds out of South-Southeast ~ 8 mph, shifting to East 10-12 mph at midday, Employees worked 9:30 AM – 1:45 PM

Conditions: 2-24: ~4.8 m/s out of the E to ESE (115 degrees). Employees worked 9:30 AM – 2:00 PM

Percent Organic Matter for Settled Solids at Lomax Deep COM from 2/21/23 to 2/24/23



The sediment trap set at the “North” position was lost due to failure of equipment. The East trap had the lowest sediment collection but the highest % OM, in keeping with the previous patter. However, South and East had % OM here in proportion to their sediment collections. The multiday deployment may have captured a variety of conditions altering that pattern.

Sediment Simulation Model

The measured characteristics of sediment plumes were summarized in a table (Summary Results Table; see end of results section) which served as our Sediment Simulation Model. The Sediment Simulation Model provided evolving guidance for the seagrass experiments that follow below.

Sediment disturbance experiments at seagrass beds were usually within or near the range of measures observed or simulated at shallow and deep COM sites. Experiments at seagrass sites attempted to use this guidance as “middle values” in order to bracket these values to represent the range of potential sediment impacts.

Conditions and Sediment Plume Measurements at Seagrass Sites

Study Site Scouting and Selection

Maps of seagrass occurrence and water depth were used to identify areas with potential to find suitable monotypic *Halodule wrightii* seagrass beds of large size and consistent depth, ideally with a southeastern flanking unvegetated edge or strip. Several scouting trips were used to visit 12 potential sites December 2021 – April 2022 to examine the seagrass beds for acceptable characteristics to conduct our experimental design. Sites are indicated by an aerial image in Methods. Two sites were selected, one with more muddy sediments (Turtle Bayou) and the other was more sandy (San Jose Island). These sites were pre-sampled and prepared for the experimental design as descried elsewhere in this document.

Turtle Bayou Site Data and Abandonment

The Turtle Bayou site was chosen for its large *Halodule wrightii*-dominant seagrass bed with consistent bottom characteristics and a southeast unvegetated edge for generating sediment plumes. The sediments were very muddy, and this site provided a good contrast to the relatively sandy San Jose site. The southeast edge also paralleled a deeper channel that connected to Aransas Bay. We predicted that prevailing southeast winds and incoming tides together would produce a current flow to carry sediment plumes along our track layout.

We setup our track layout according to the standard map, performed pre-experiment sampling (seagrasses, invertebrates, physical measurements and UAS imagery), and began to apply sediment disturbances on multiple occasions in July and August. We encountered great difficulties with hydrology at the site, which manifested as sediment plumes not travelling along the tracks. Rather, to our surprise, the sediment plumes either did not move at all, or else they actually travelled the opposite direction into the channel. This occurred even when the wind was out of the southeast at 17 mph and the tide was simultaneously incoming, both of which should have pushed the sediment plumes over the shallow seagrasses. The wind-driven waves clearly moved in the expected direction, along with floating pieces of seagrass. But the plumes were moving in a counter-current out of the tracks and backwards into the channel. We tried multiple adaptive strategies to make this site suitable for our study, but none were successful. It was necessary to abandon this as a study site and focus instead on the San Jose seagrass site.

The pictures below were captured from video on the aforementioned day with strong southeast wind and incoming tide. Both light and heavy disturbances are being applied (1 vs 2 walkers). The sediment was expected to move to upper right over the seagrass, but it clearly moved backwards despite an incoming tide.

Turtle Bayou (3 Tracks; Light and Heavy Disturbances near start)

Turtle Bayou (3 Tracks; Light and Heavy Disturbances)

Red Arrows: Approx wind direction

Yellow: Approx current direction

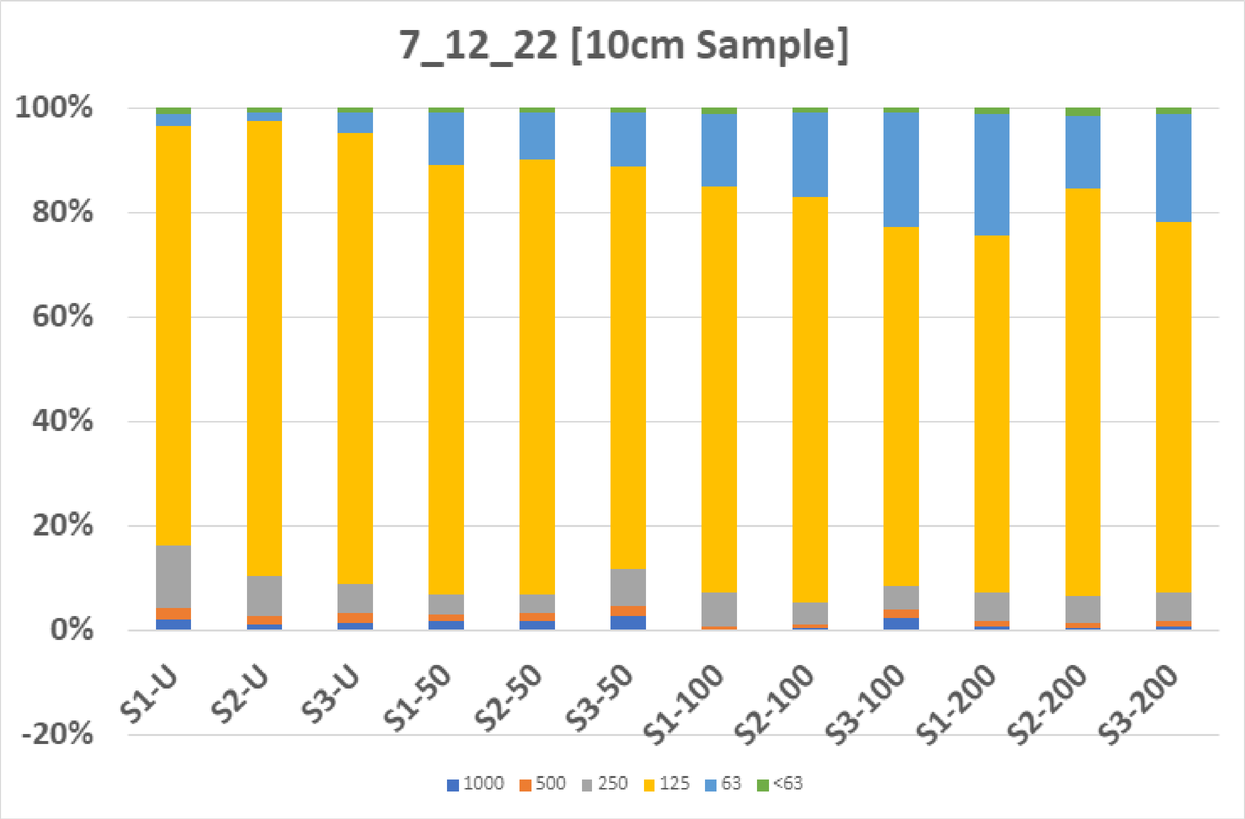
Blue: Direction of sediment plumes

San Jose Island Site Data

Sediment Cores

The seagrass study sites were characterized by pre-experiment sampling (seagrasses, invertebrates, physical measurements and UAS imagery), which included 10 cm deep core samples to characterize the sediments across the study site. Sediment samples were processed to characterize grain size, depicted in the figure below. Grain size varied slightly across the site in a pattern that reflected the depth gradient. Grain proportion in the 63 –125 micron size class increased from <5% to ~ 20% moving from the track baseline towards the 200 ft boundary at the deeper edge of the study site. Grain proportions larger than 125 microns decreased accordingly. This difference occurred along the track length and there were no clear differences between the different tracks which represented different treatments. This eliminates one potential confounding factor for comparisons between tracks (sediment treatments).

Sediment Core Grain Size Composition at San Jose Seagrass Study Site



Note increasing proportions of fine-grained material (63 µ, blue) at increasing distance from baseline and depth of water.

Disturbance Log and Documentation of Sediment Disturbances at Seagrass Sites

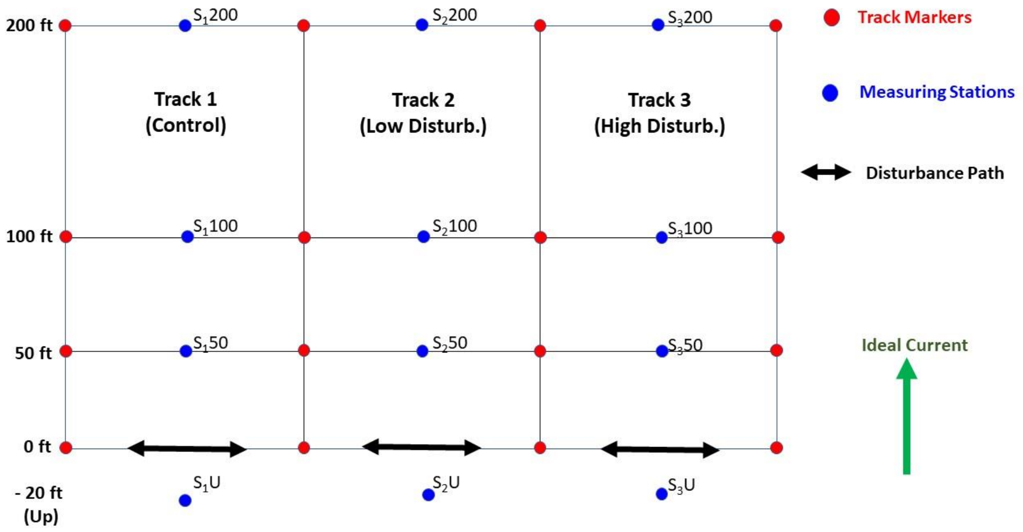
The table below represents part of the Disturbance Log that was kept to document important metadata regarding sampling and sediment disturbances applied at the San Jose seagrass study site. Only data for this study site is depicted here. The unit p-m (person-minutes) was used to describe disturbance intensity. Track 3 (Heavy Disturbance) received a total of 811 p-m of disturbance, compared to Track 2 (Light) which received 455 p-m and Track 1 (Control, mostly undisturbed) which received only 30 p-m.

Date	Disturbance (P-M)	Wind Direction	Wind Speed Avg	Tide	Current
6/3/2022	Track 2 **(15 P-M)**	E	4.75 m/s	NA	NA
	Track 3 **(45 P-M)**				
7/14/2022	Track 2 **(5 P-M)**	SSE	5.2-6.2 m/s	NA	NA
	Track 3 **(5 P-M)**				
	Track 2 **(10 P-M)**				
	Track 3 **(20 P-M)**				
	Track 2 **(20 P-M)**				
	Track 3 **(40 P-M)**				
7/18/2022	Track 2 **(5 P-M)**	SE-SSE	12-15mph	High Tide and flat, becoming outgoing	Current towards NW, then variable as tide becomes outgoing
	Track 3 **(10 P-M)**				
	Track 2 **(20 P-M)**				
	Track 3 **(40 P-M)**				
8/27/2022	Track 3 **(10 P-M)**	SE-SSE	3.7-4.1 m/s	Incoming Tide	Current towards N - NE
	Track 2 **(5 P-M)**				
	Track 3 **(40 P-M)**				
	Track 2 **(20 P-M)**				
	Track 3 **(80 P-M)**				
	Track 2 **(40 P-M)**				
9/10/2022	Tracks 2 & 3 **(40 P-M)**	ENE	0-2.1 m/s	High and Flat	Current highly variable due to shipping activities; Observed reversals from Northward to Southerly
	Tracks 2 & 3 **(80 P-M)**				
9/17/2022	Track 3 **(40 P-M)**	SSE to SE	2.9 m/s	High & Outgoing	Current towards SW
9/25/2022	Track 3 **(46 P-M)**	W	2.6 m/s	High & Outgoing	Current towards S – SW, perpendicular to track and slightly backwards
10/1/2022	Track 3A **(120 P-M)** 3B **(20 P-M)** & 3C **(10 P-M)**; Track 2A **(60 P-M)**	NNE to NE	4.5 m/s	High & Outgoing	Current towards S – SW; Strong outgoing tide and NE to NNE winds together moving current perpendicular to track

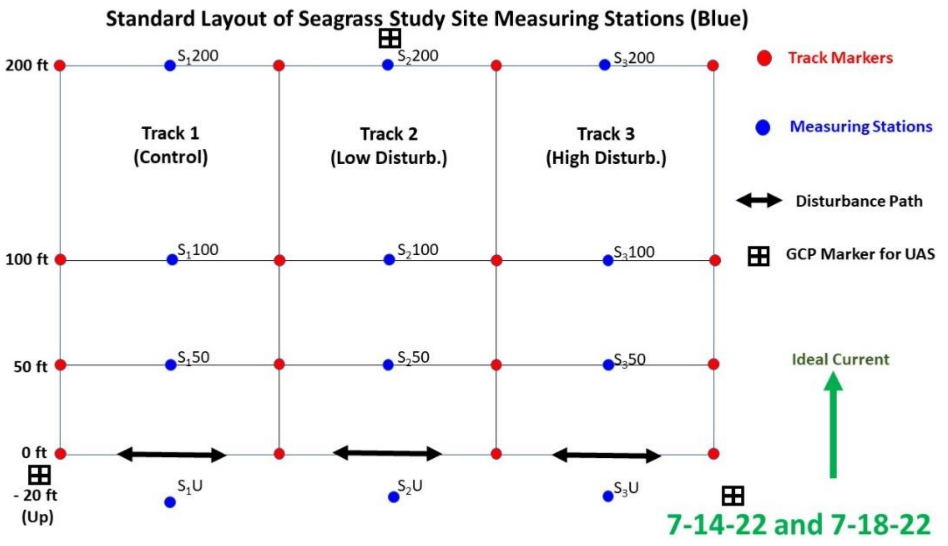
10/5/2022	Track 3 **(30 P-M (20P-M per 50 ft))**	NE to ENE (51 deg)	4 m/s	Medium High & Outgoing	Low current, Current towards SE for 1st 10 min disturbance; Then North Westerly Current for 2nd disturbance
10/15/2022	Track 2 **(45P-M)** Track 3 **(90P-M)**	SSE 140 to 160 degrees	3.3 m/s	High and slack	Little to no current
11/2/2022	Track 3 **(185 P-M)**	NE 40 to 45 degrees	6.2 m/s	NA	Current towards S
11/9/2022	Track 2 **(140 P-M)**	E	2.7 m/s	Incoming Tide from S-SW	Current towards NE

Total P-M:	**Track 1: 30 P-M**
	Track 2: 455 P-M
	Track 3: 811 P-M

It was necessary to adapt the sediment disturbance experiments to existing current conditions. On multiple occasions the conditions in early morning changed during the late morning, and a completed measuring pole array had to be removed and setup differently. Below is a seagrass study site map that shows the initial experimental design based on the assumption of predominant water currents out of the southeast would be driven by the predominant southeast winds. Following that is a series of modified maps indicating how sediment disturbances were actually applied along with the actual locations of the measuring stations adapted for the given conditions.

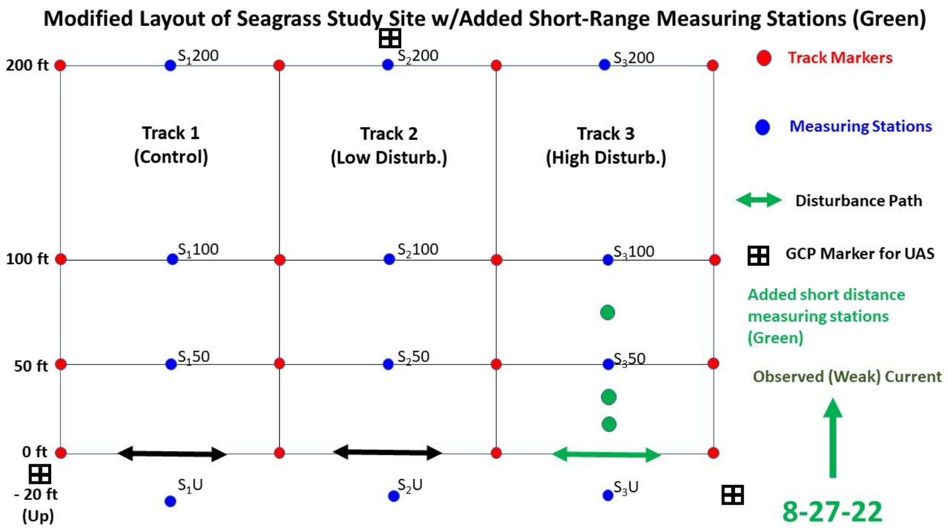


Standard configuration for wind driven currents out of the southeast



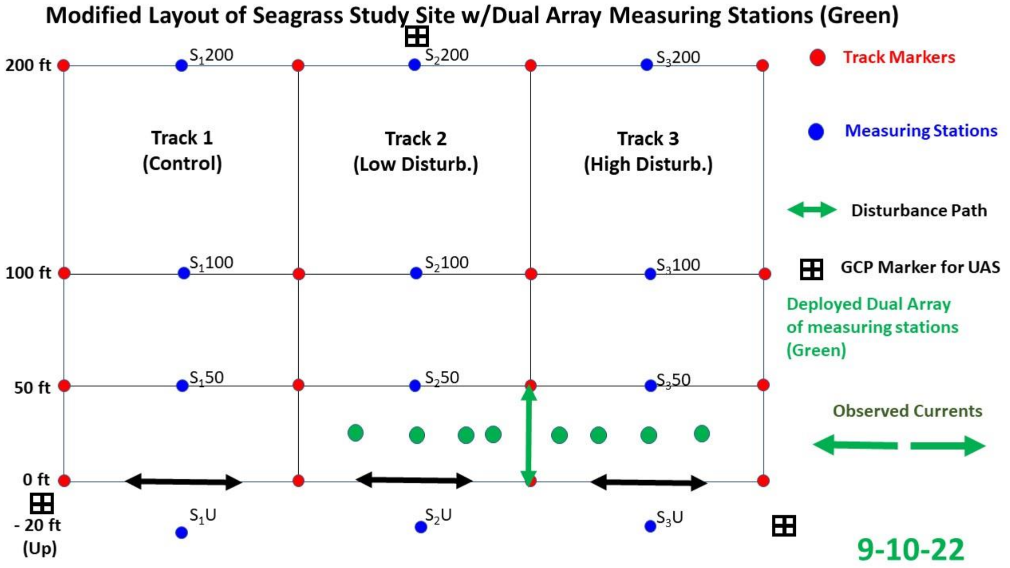
Currents were only weak, so sediment moved in correct direction, but not very far (barely reached the 50 ft pole).

Adapted by adding light logger poles and sediment traps at shorter distances (added 20, 35 and 75 ft stations)



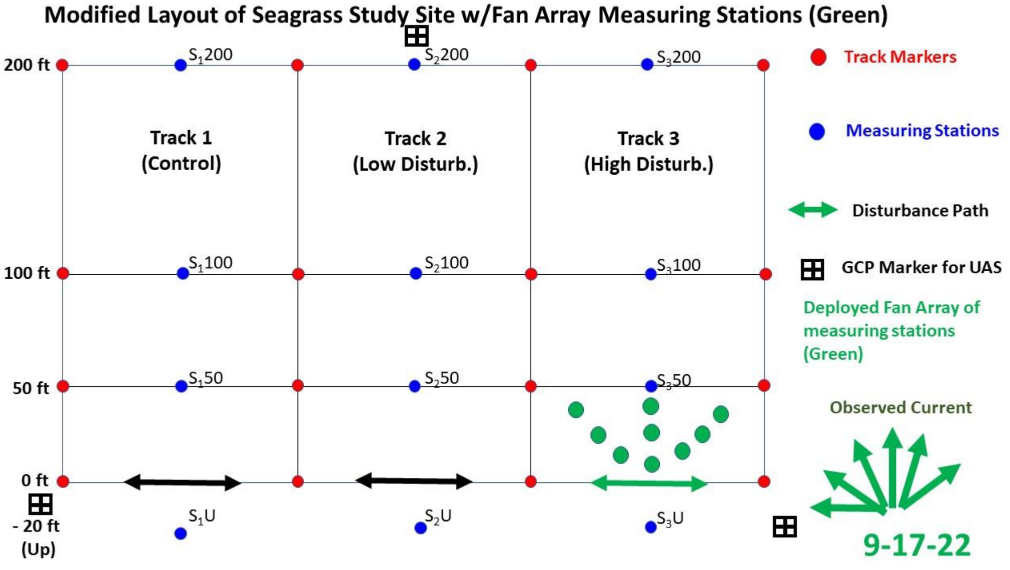
We frequently observed strongly shifting currents and seagrass blades laying in one direction, but shortly later shifting their orientation, often along a NE to SW axis (see pictures below). To document that, a dual array of measuring stations was setup as illustrated below in green. A disturbance was applied in the center to track direction of sediment plume movement (see below). Note, we learned this was caused by large oil tankers passing through the Aransas Pass Ship Channel 5 miles to the South.

Modified Layout of Seagrass Study Site w/Dual Array Measuring Stations (Green)



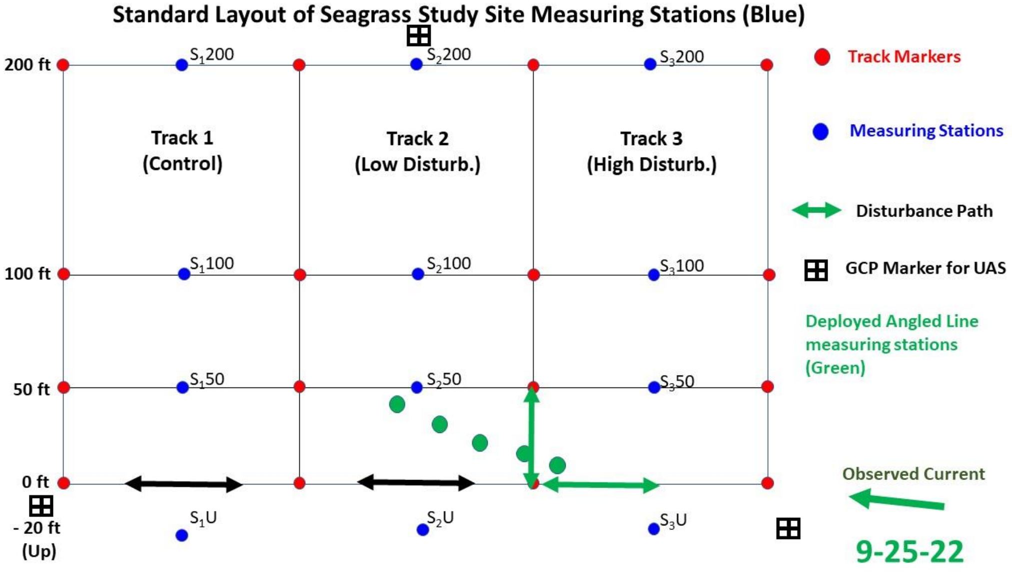
Sometimes sediment plume started in one direction towards our loggers, and then shifted, missing the logger stations (see pictures below). To avoid missing the sediment plume, we arranged “Arrays” of measuring stations fanning out in different directions to increase the probability of measuring the plume.

Modified Layout of Seagrass Study Site w/Fan Array Measuring Stations (Green)



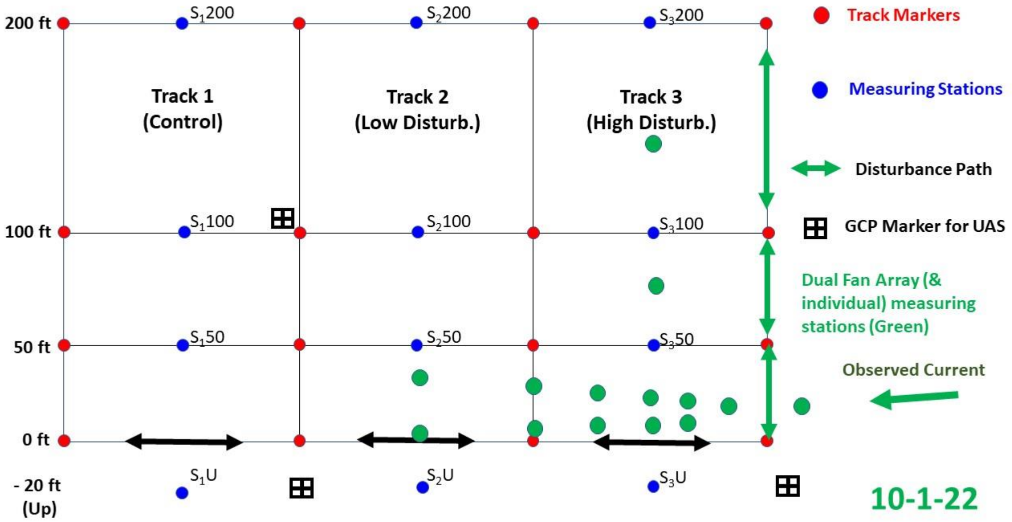
For currents that were off-axis relative to our tracks, we used an angled pole array and adjusted the baseline of sediment plume generation to be up-current.

Standard Layout of Seagrass Study Site Measuring Stations (Blue)

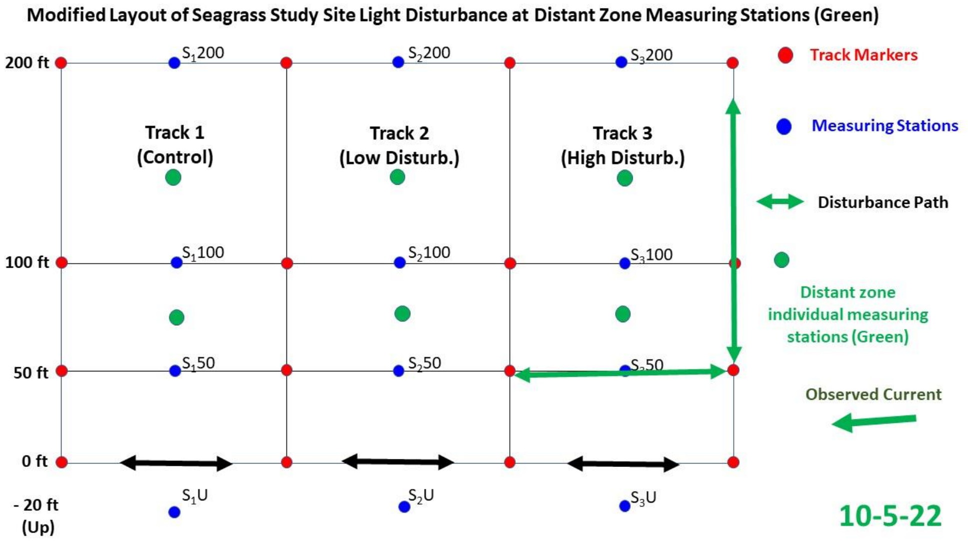


We learned that a northeast wind in conjunction with an outgoing tide produced a current that was almost exactly perpendicular to our track orientations. We adapted by applying sediment disturbance on the NE end of the study site directly upstream from Track 3 (heavy disturbance treatment). This arrangement resulted in a near perfect movement of the sediment plume across the 3 tracks in the 50 ft Zone “A”, moving from Track 3 to Track 2 to Track 1 (and beyond 300 ft). This was captured with a dual fan array as depicted below. The net effect was a heavy disturbance in Tr 3-50, a medium disturbance in Tr2-50 and a light disturbance in Tr1-50. After two disturbances in Zone A, 2 lighter disturbances were applied in Zones B and C, from the NE edge. This achieved the same disturbance intensity differences as prescribed in the original plan.

Modified Layout of Seagrass Study Site w/Dual Fan Array Measuring Stations (Green)



Similar to the adjustment described above, disturbances applications were adjusted to target Zones B and C as shown below.



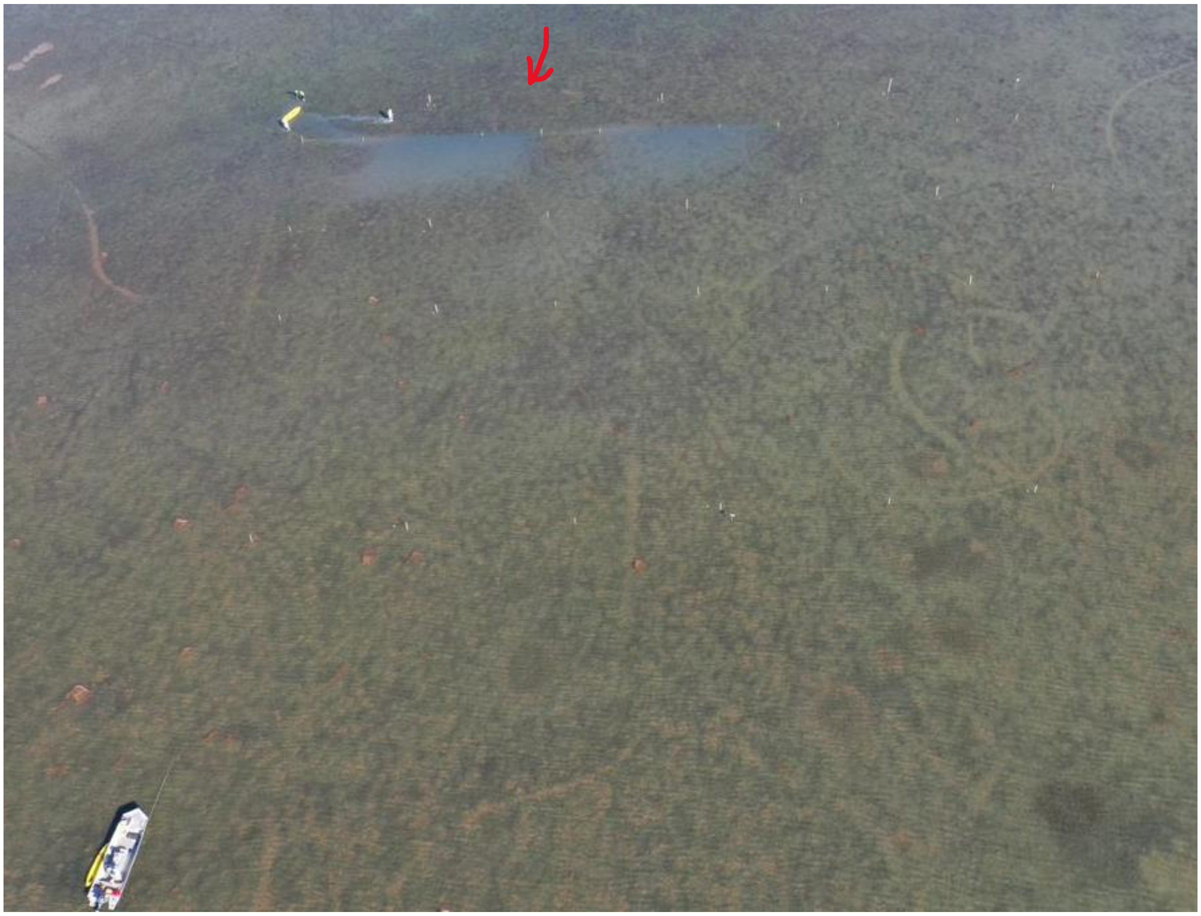
Sediment Plume Picture and Video Observations at San Jose Seagrass Site

San Jose 7-14-22 (Tracks 2 & 3; Light and Heavy Disturbances)

Note that current moved backwards and to East, despite strong winds out of the SE (parallel to tracks).

San Jose (UAS; 3 Tracks; Light and Heavy Disturbances; Near start)

San Jose (UAS; 3 Tracks; Light and Heavy Disturbances: After ending)



Note that sediment plume has not yet reached the 50 ft pole after 20 minutes.

San Jose (UAS; 3 Tracks; Light and Heavy Disturbances; 30 min after ending)

Note that sediment plume is widely dissipating, and just barely at the 50 ft pole. This type of observation led to setting up poles at shorter distances from the sediment source.

San Jose 9-10-22: Disturbance between dual Southwest and Northeast Arrays aligned NE to SW (eg perpendicular to Tracks)

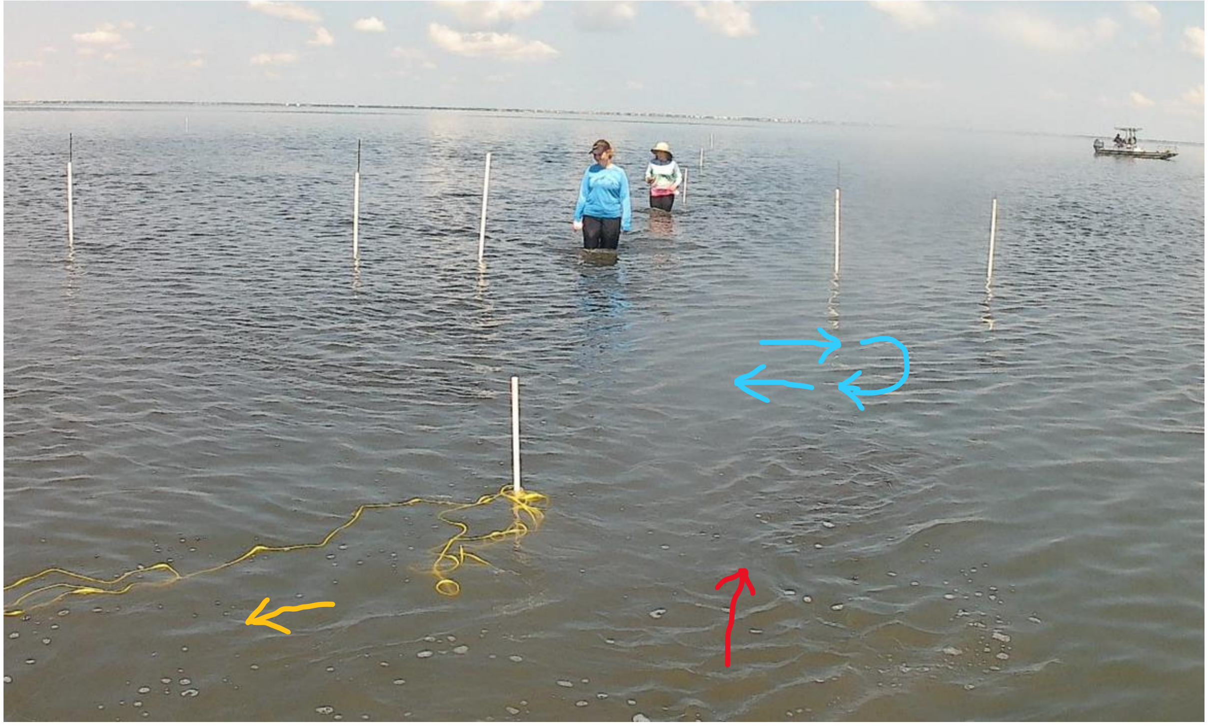
Attempts to measure shifting currents led to this experimental design; Nearly no wind; Half of measurement array faced to Northeast and half faced to Southwest, with a disturbance track between the two. Applied a heavy disturbance (80 p-m) and photographed.

San Jose 9-10-22 (Sediment plume moving over Measuring Stations to Northeast)

Note that plume is moving NE over this half of array; Not pictured is absence of movement across other SW half. The first 3 light loggers and sediment traps (out to 20 ft) are not visible due to the plume.

However, after approximately 10 minutes, the sediment plume reversed course and began moving to the SW, eventually uncovering the NE loggers and covering the SW array of loggers.

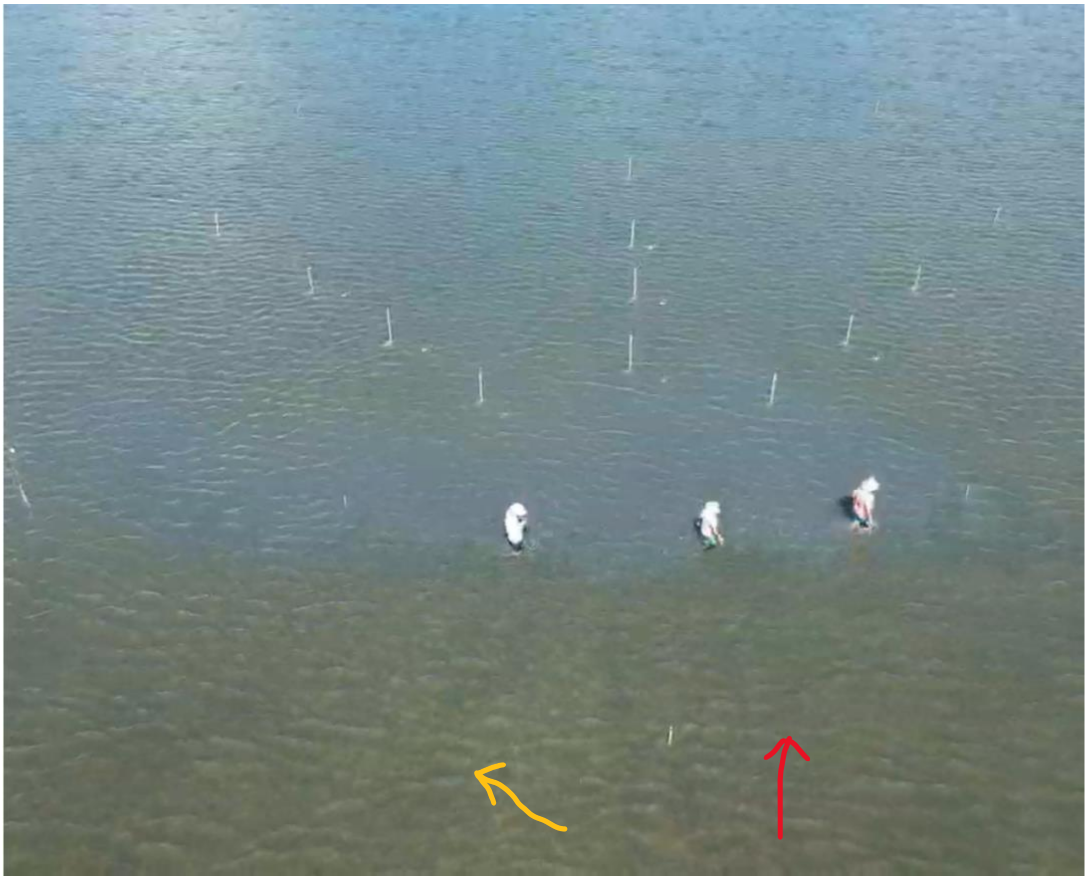
See this “Flip-Flop” in picture below.



(Light winds, slight outgoing tide, tankers thru ship channel: Observed **"flipflop"** of SG and sediment plume)

"Flip-Flop" is an effect of large-displacement tankers moving through Aransas Pass 5 miles to South.

San Jose 9-17-22 Track 3 Heavy Disturbance w/Fan Array of Poles (series of photos at 4 different times early → late in a 40 min Disturbance). Note how the plume changes course, avoiding most of the logger poles.



Plume has reached the first pole on right side of array, and approaching first pole in center.

Plume has reached first pole in all 3 directions.

Plume begins to pull away from 1st poles in all 3 directions and moves off towards the left.

Most of plume has moved another direction away from poles.

San Jose 10-1-22 Track 3 Heavy Disturbance w/Dual Fan Array of Poles Oriented for NE Current

With wind and tide oriented in synchrony to produce a strong current perpendicular to the tracks, the measuring array was adapted as described in a site map above. Three Disturbances were applied along the NE side of the site and currents swept a sediment plume across all 3 tracks. The 2nd disturbance (2 people x 30 min = 60 P-M) is depicted at different times in UAS imagery below. Note that the plume encompassed entirely the Zone “A” (0-50 ft) of Tracks 3, 2 and 1. Note how the plume progresses across the tracks and eventually exceeds 300 ft of travel post-application as it begins to clear from near the source. Numerous example measurements were derived from this experiment in the following figures.

10-1 San Jose Dist #2 (10:16 – 10:36)
Video # 0835 at **10:18:30 AM (2.5 min Very Early in Dist #2)**

Approaching 20 ft

10-1 San Jose Dist #2 (10:16 – 10:36)
Video # 0835 at **10:21 AM (5 min Early in Dist #2)**

At 35 ft

10-1 San Jose Dist #2 (10:16 – 10:36)
Video # 0836 at **~10:23:30 AM (8 min Middle of Dist #2)**

Approaching 100 ft

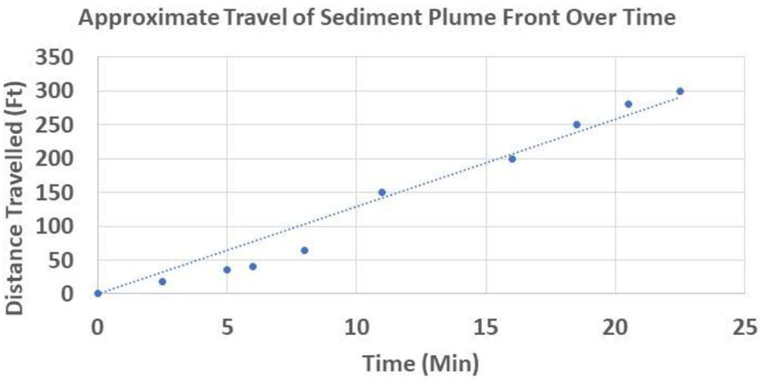
10-1 San Jose Dist #2 (10:16 – 10:36)
Video # 0837 at **~10:27 AM (~11 min Near Middle of Dist #2)**

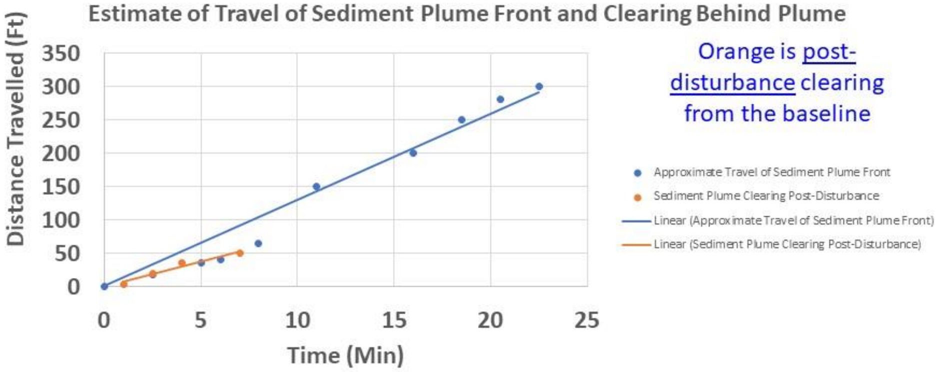
Approaching 150 ft

10-1 San Jose Dist #2 (10:16 – 10:36)
Video # 0838 **10:34:30 AM, 2.5 min into video (18.5 min Shortly before end of Dist #2)**

At approx. 250 ft

Observation of the imagery allowed analysis of the movement of the sediment plume with time:


Approximate Travel of Sediment Plume Front Over Time

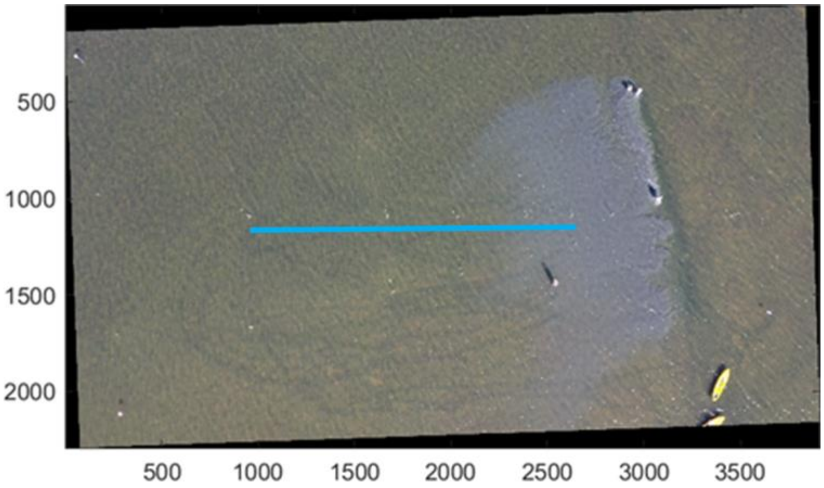

Estimate of Travel of Sediment Plume Front and Clearing Behind Plume

Orange is <u>post-disturbance</u> clearing from the baseline

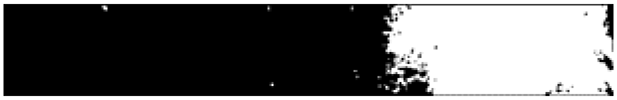
Observation of plume front over time estimated plume travel time of ~ 10 min per 100 ft. Slower but somewhat similar rate of clearing behind the plume following disturbance.

Image Analysis of Sediment Plume Movements at Seagrass Sites

For plume movement analysis, first each analyzed image frame was rotated to align the upper marker posts so that they were horizontal in the image frame. Next, region of interest (ROI) was selected as a rectangular area bounded by the midpoint or intersection of two 200 foot marker posts on the left, the upper marker posts on top, midway between upstream marker post and 20 foot marker post on the right and 100 foot marker post at the bottom.



The image was then smoothed (mean filtered) multiple times to remove the effects of glint. The image converted into HSV color space, and thresholded to obtain the binary image that represented the plume area. In the image below, black represents background (pixel value of 0) and white represents plume (pixel value of 1).

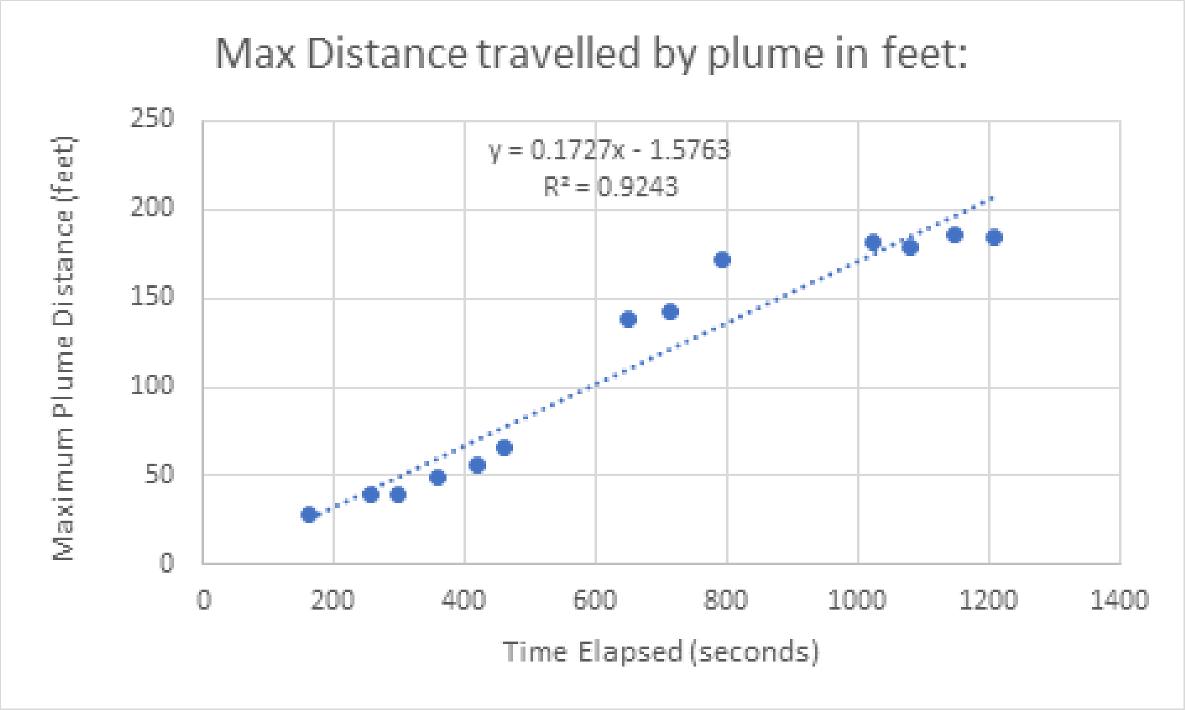


To find the estimate distance traveled by the generated plume, each of the row pixels was summed and maximum row sum was identified. The pixels were then calibrated to world coordinates from the marker posts. This allowed independence from UAV hight which affected the scale of the viewing area.

The above process was repeated for different image frames with different time stamps. Maximum distance traveled by plume at a given time was determined after pixel calibration and recorded for each image vs time elapsed.

Plume flow is estimated from the gradient in the following graph based on distance traveled in feet per second. After converting the time elapsed in seconds to time elapsed in minutes, the **estimated plume flow rate is found as 10.2 feet per minute**.

Image-derived sediment plume travel estimation:



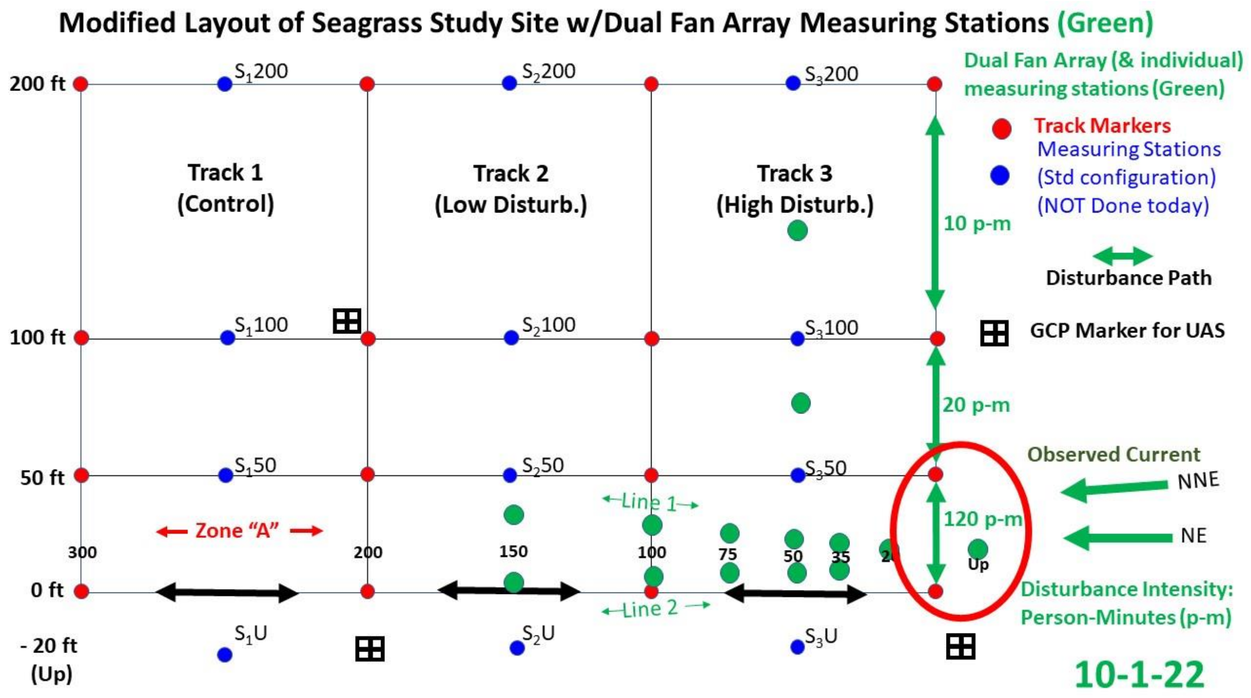
Estimate average of 10.2 ft/min rate of plume travel over course of Disturbance 2

1. **Sediment Plume Physical Measures**
   a. **Light Logger Data**

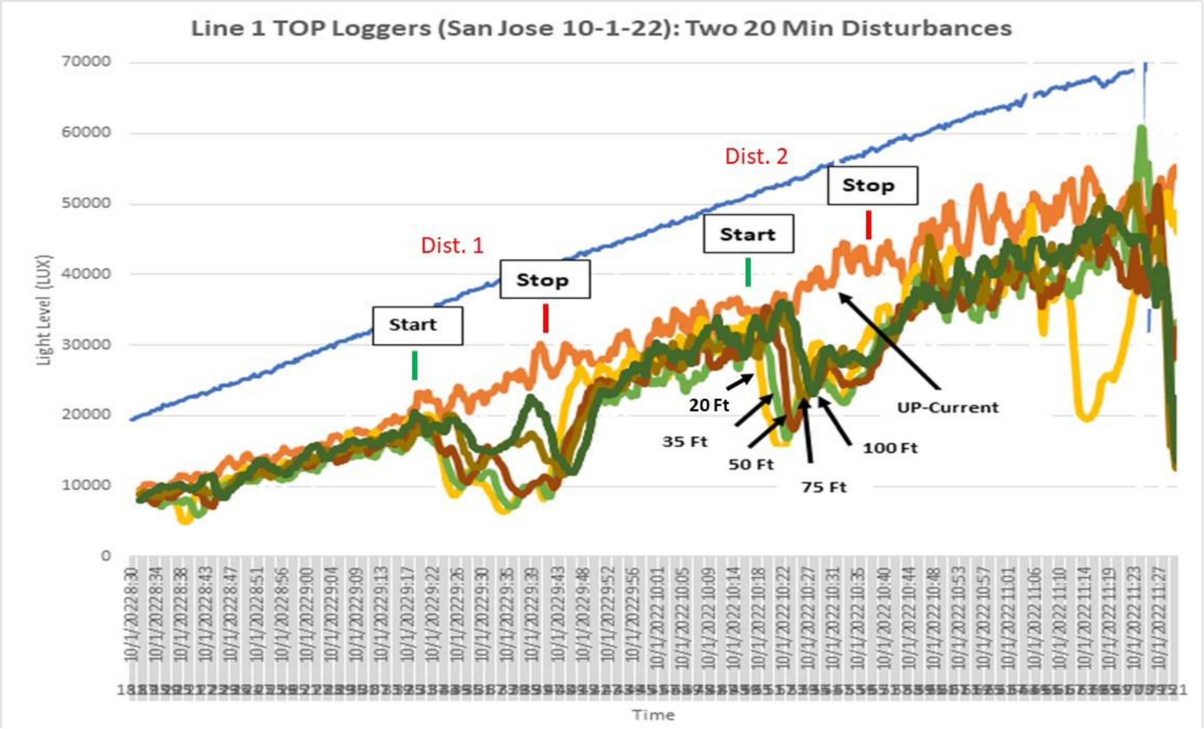
An example of light logger analysis of sediment plume travel is provided in the figures below. A variety of approaches were explored for generating sediment plume metrics from light logger data.

<u>10-1-22 San Jose Experiment as example of complete analysis</u>:

Experimental Design (Disturbances 1 and 2):



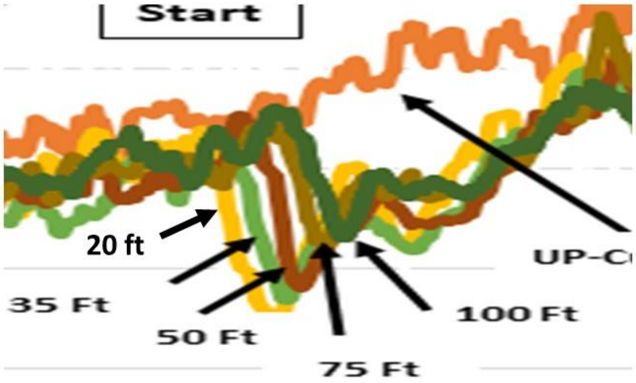
Example Light Logger Data: Raw data is the measured light level over time.

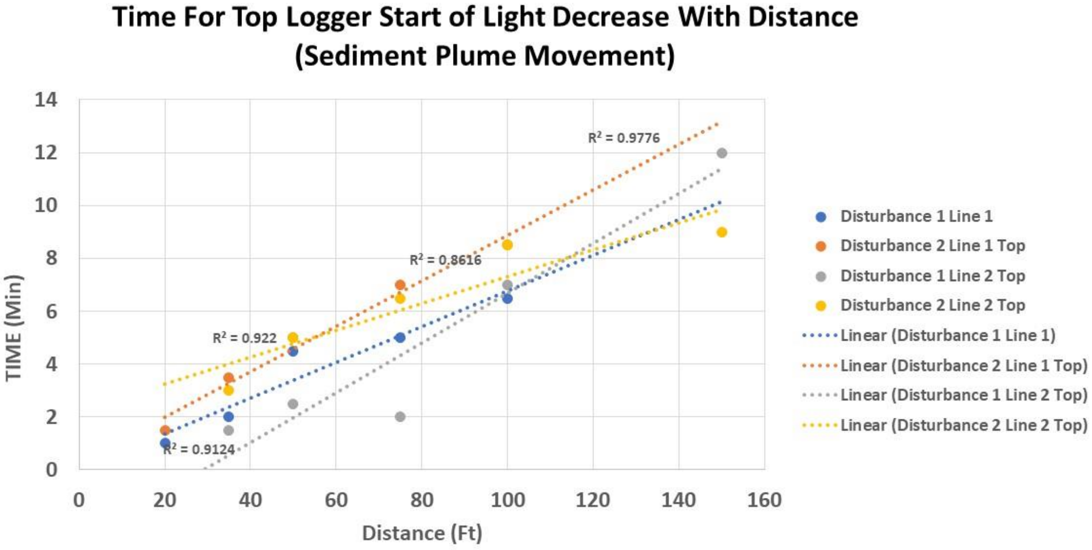


For Disturbance 2:

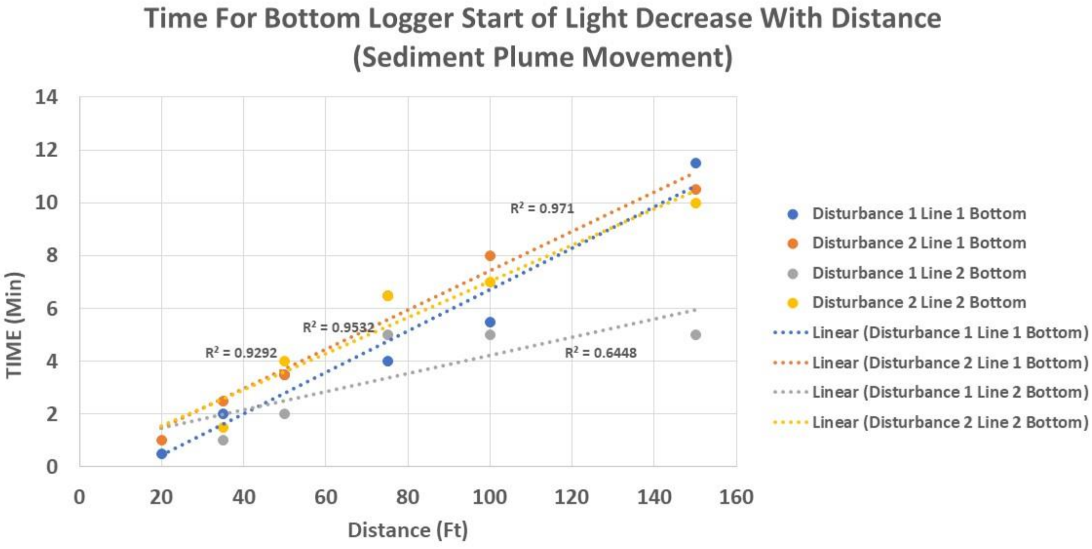
Order of Light Decrease from Light Loggers

20 ft → 35 ft → 50 ft → 75 ft → 100 ft

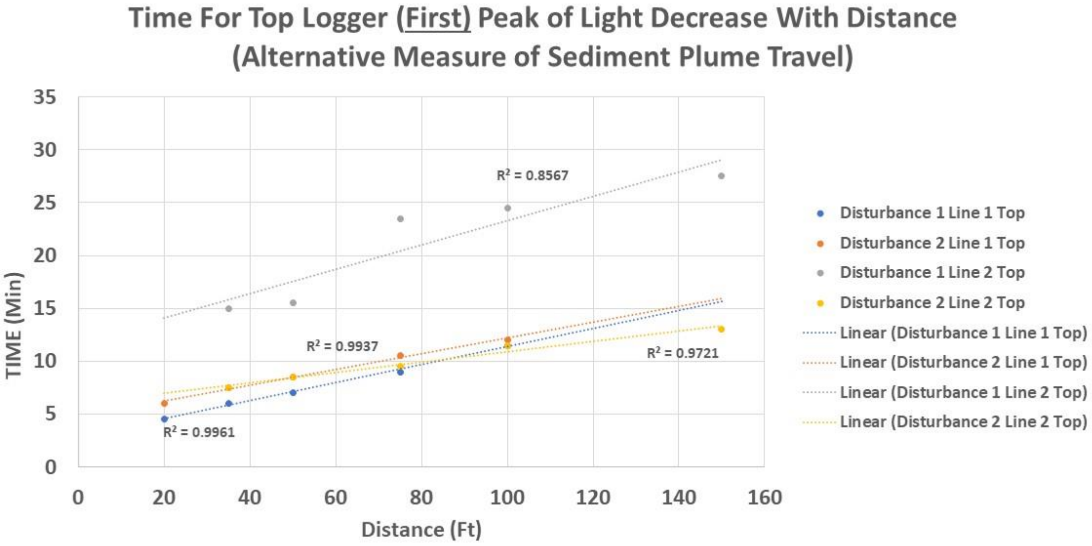




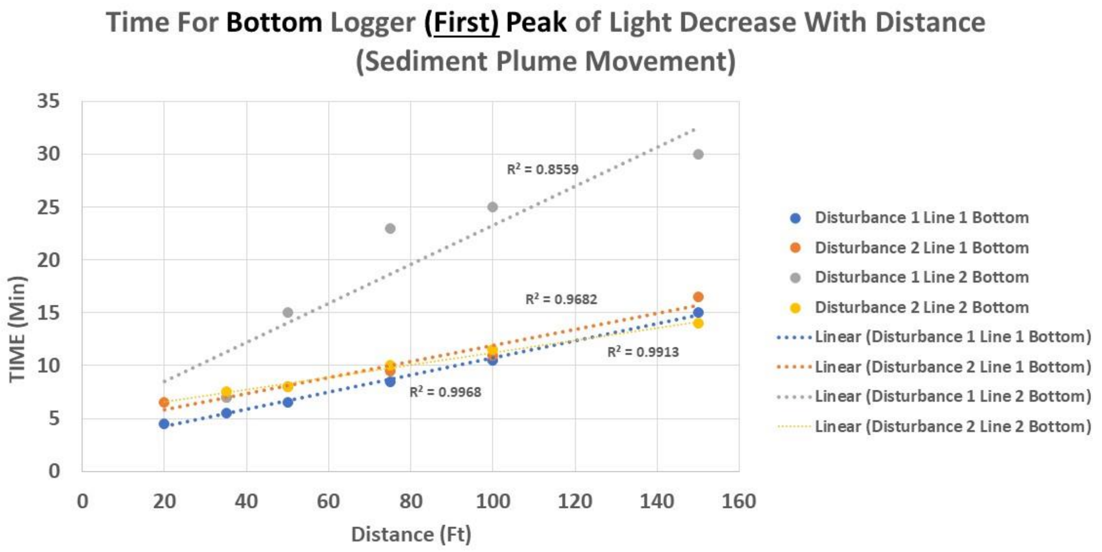
Initial light decrease from two lines of loggers during 2 separate disturbances show similar rates of sediment plume movement



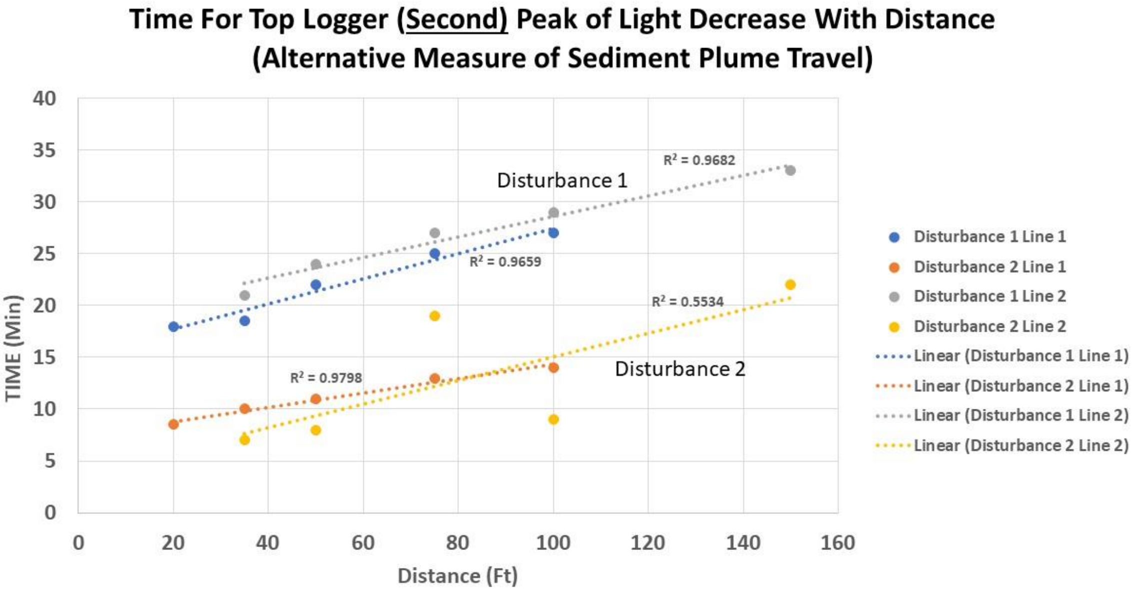
Similar observations at the lower of the 2 light loggers ("Bottom"), although a Line 1 logger at the farthest distance (150 ft) had divergent results.



Alternative measure is to use the time of peak light decrease. However, this could be impacted by other factors, and appeared less robust.

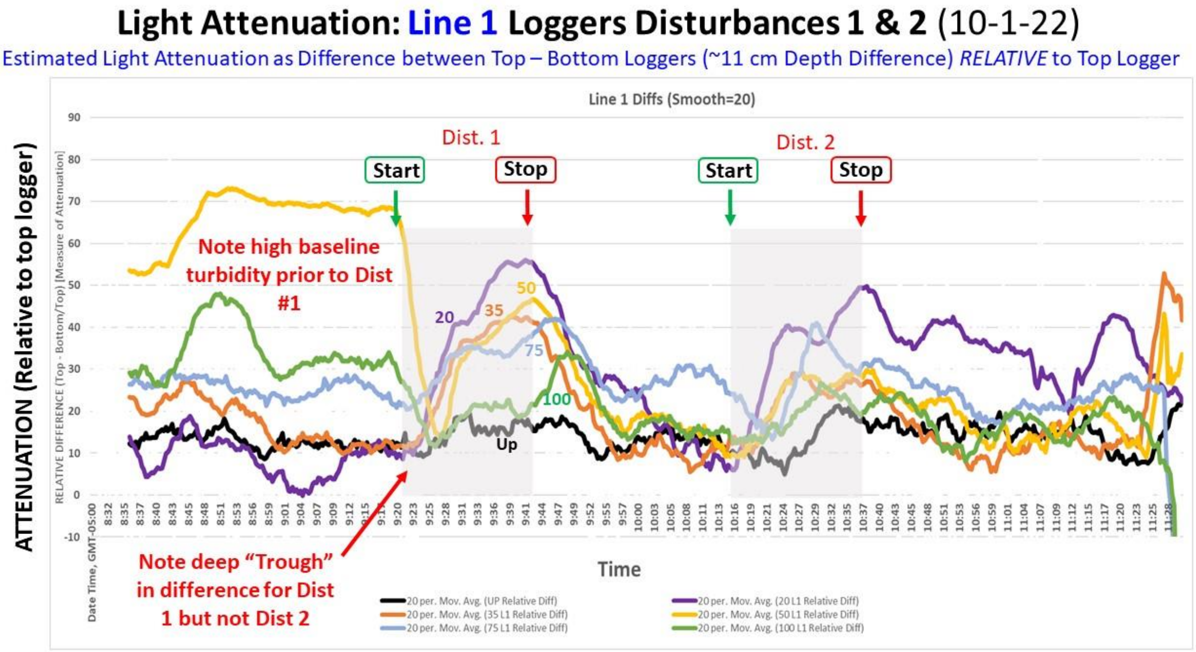


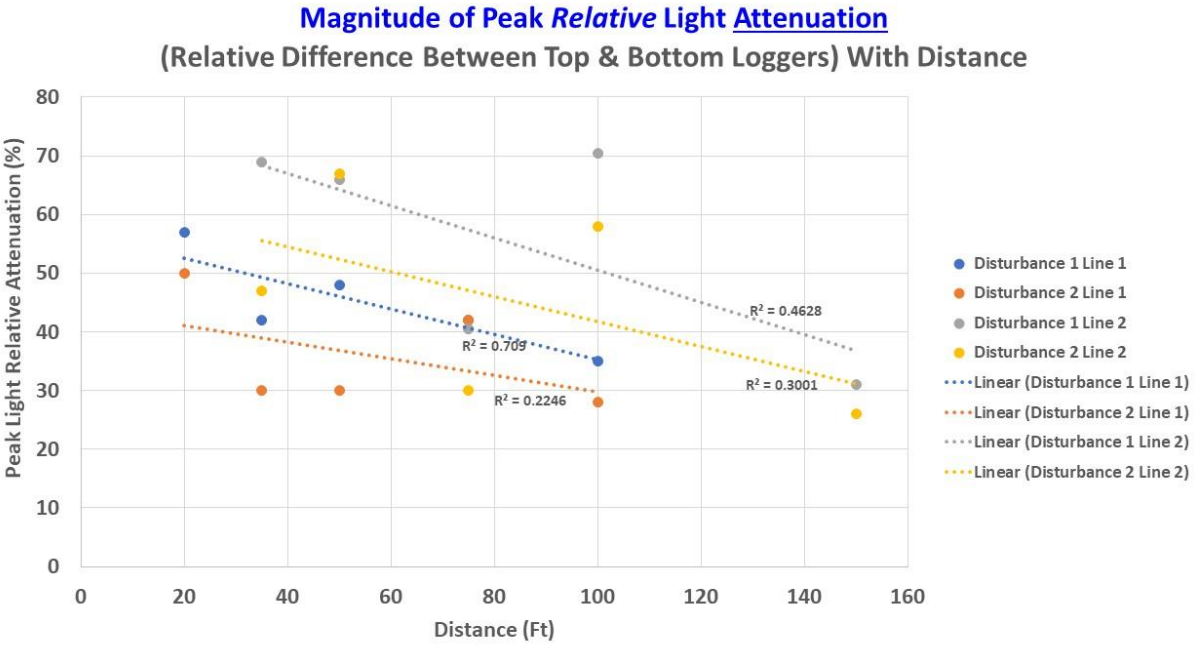
Alternative measure is to use the time of peak light decrease. However, this could be impacted by other factors, and appeared less robust.



Alternative measure to use the timing of the second attenuation peak as a measure of the sediment plume disturbance was less robust and derived different relationships for the 2 disturbances.

Alternative measure is to use the Relative Difference between the two light loggers as a measure of both timing and intensity of the sediment plume disturbance. This metric was difficult to measure in this test case.

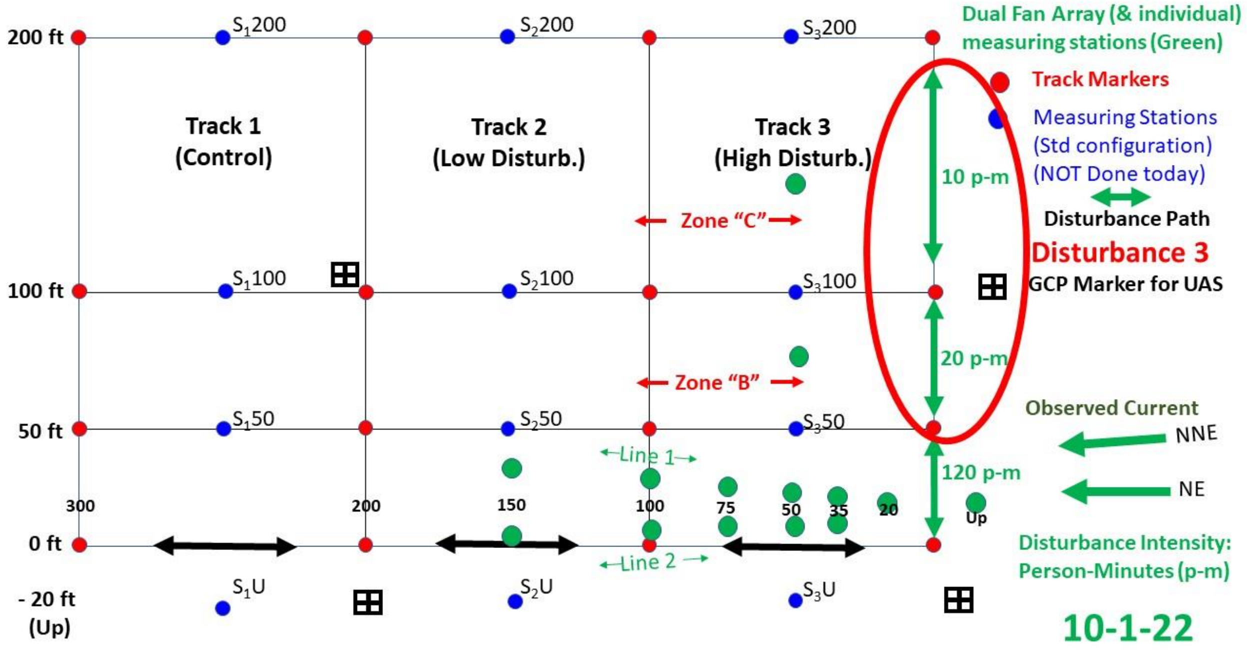

Light Attenuation: Line 1 Loggers Disturbances 1 & 2 (10-1-22)
Estimated Light Attenuation as Difference between Top – Bottom Loggers (~11 cm Depth Difference) *RELATIVE* to Top Logger


Magnitude of Peak *Relative* Light Attenuation
(Relative Difference Between Top & Bottom Loggers) With Distance

An additional disturbance on 10-1-22 was applied to produce sediment plumes of lesser intensity at Tracks 2 and 3 respectively.

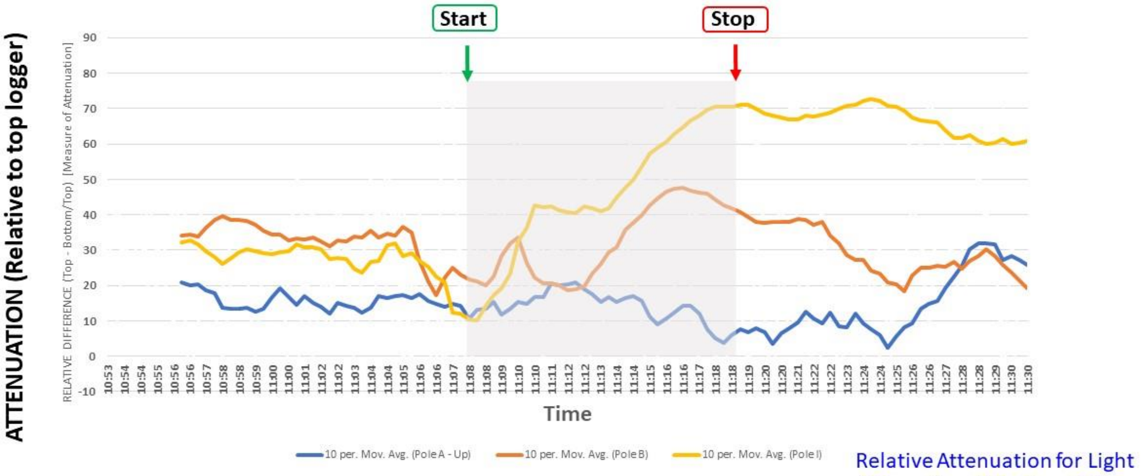
DISTURBANCE 3 targeted more-distant areas of Track 3 (Zones "B" and "C") with medium and light disturbances, respectively:

Modified Layout of Seagrass Study Site w/Dual Fan Array Measuring Stations (Green)



Light Attenuation: Disturbance 3 (2 Loggers @ 50 ft)

Estimated Light Attenuation as Difference between Top – Bottom Loggers (~11 cm Depth Difference) *RELATIVE* to Top Logger



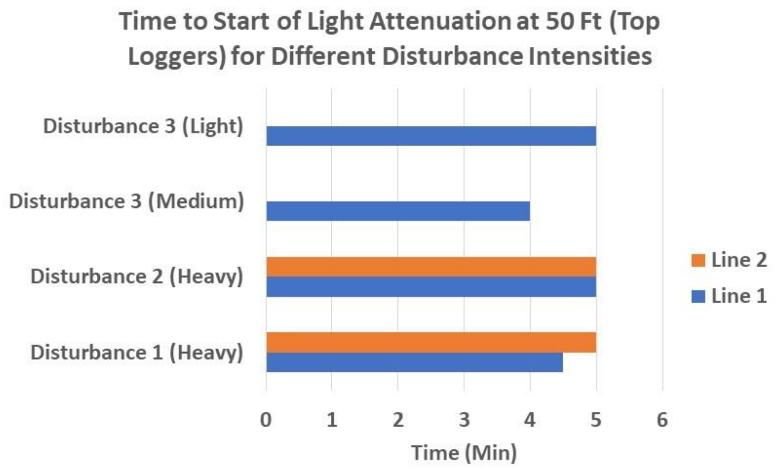
Relative Attenuation for Light (10 p-m) and Medium (20 p-m) Disturbances at 50 ft

Do light attenuation metrics vary w/intensity of disturbance ?
Yes for light and medium disturbances
Compare with heavy disturbances 1 & 2

The 10-1-22 experiment enabled a direct comparison of disturbances of Heavy, medium and light intensities when performed under nearly identical conditions.

Comparison of rate of movement for different intensities (Time to Start of Light Decrease at the 50 Ft Pole):

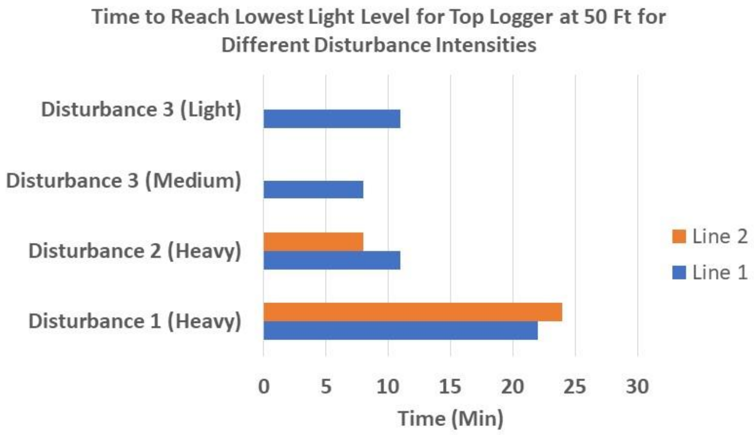
Comparison of Relative Attenuation for Light (10 p-m), Medium (20 p-m) and Heavy (60 p-m) Disturbances at 50 ft (10-1-22 San Jose Expt)



Rate of sediment plume movement was independent of the Disturbance intensity

Comparison of rate of movement for different intensities (Time to Reach Lowest Light Level at the 50 ft Pole):

**Comparison of Relative Attenuation for Light (10 p-m), Medium (20 p-m)
and Heavy (60 p-m) Disturbances at 50 ft
(10-1-22 San Jose Expt)**

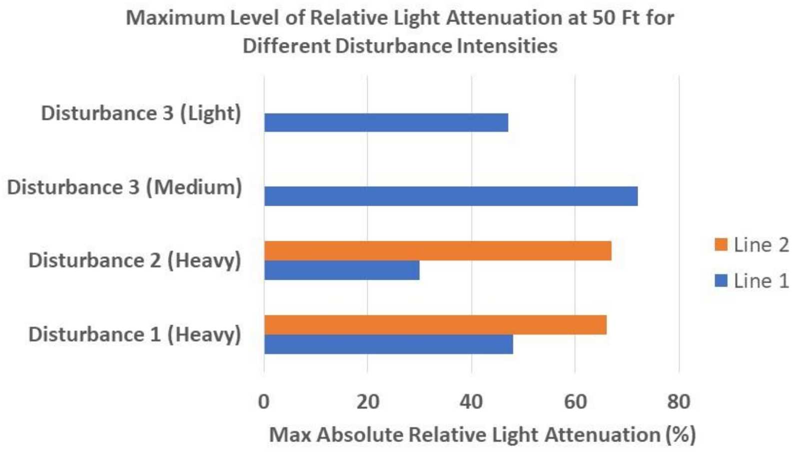


The time to reach maximum light attenuation was similar between light, medium and 1 of the two heavy disturbances. However, the first disturbance applied this day (heavy) took approximately twice as long to reach its maximum attenuation value. This suggests that the driving currents may have been different at these two different times.

Both Lines 1 and 2 were similar in the timing of the attenuation maximum.

Comparison of Degree of Relative Light Attenuation at the 50 ft Pole:

**Comparison of Relative Attenuation for Light (10 p-m), Medium (20 p-m)
and Heavy (60 p-m) Disturbances at 50 ft
(10-1-22 San Jose Expt)**

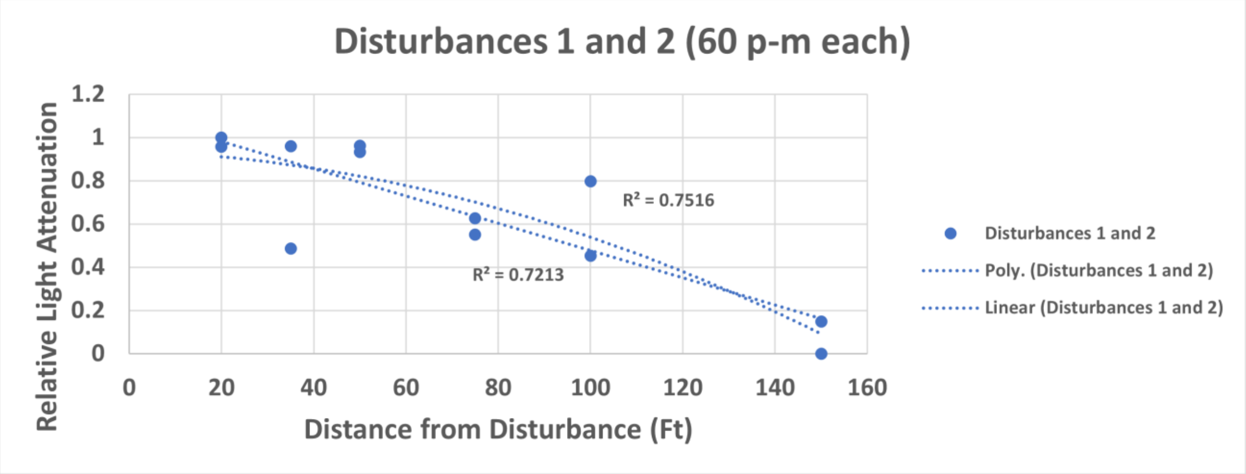


In the side by side 3rd Disturbance, maximum light attenuation shows the expected difference between light and medium disturbances.

However, we did not see further increased maximum light attenuation level at 50 ft for the two heavy disturbances, but these were done earlier in the day. Possibly a “saturation” of suspended sediments (as detectable by light loggers) ?

Also see the effect of how well the loggers align with current direction (Line 2 better aligned than line 1).

Another metric to compare light attenuation along a gradient of distance from the disturbance is the integration of the area beneath the Relative Difference Plots as a measure of the total amount of light attenuated by the passing of the sediment plume. For this, the area beneath these plots, over time and at different distances from the disturbance, was measured and corrected for ambient turbidity light attenuation at the “Up” pole. Values from the two lines of poles were averaged for each distance and plotted for the two disturbances in the graph below. Values on this plot were scaled relative to the sediment plume of greatest magnitude (observed at 20 ft in Disturbance 1) which was assigned the value of 1. A turbidity plume of ½ that magnitude (area under the graph) would have a value of 0.5 for instance.



According to this plot, the light attenuation at 150 ft was less than 20% of that measured at 20 ft. This does **not** mean that the turbidity had disappeared! The aerial imagery clearly shows the sediment plume traveling to 300 ft. The maximum attenuation at 20 ft was approximately 60% of light incident on the top logger, and at 150 ft that value was about 25%. These values are in agreement with the areas integrated beneath the Relative Difference Plots.

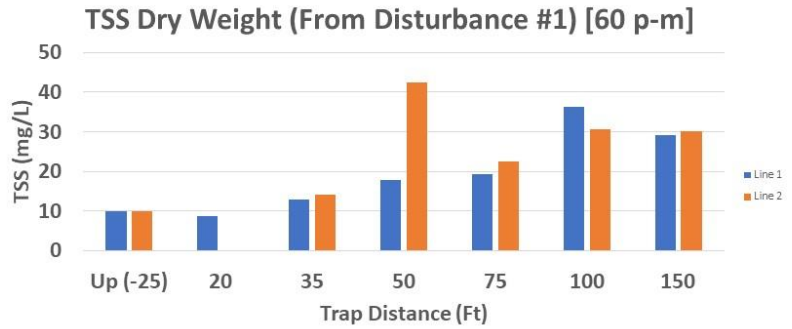
Total Suspended Solids (TSS)

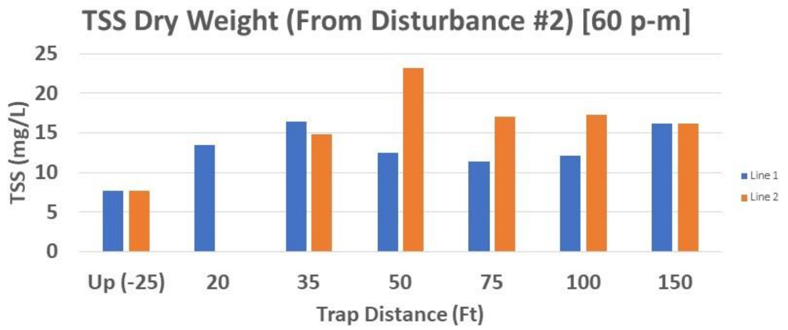
TSS samples were collected upstream and downstream immediately following sediment disturbances. However, the timing of collection varies and may impact the results depending on the speed of the current. Care must also be taken to avoid creating additional sediment disturbances while moving downstream to the collection site. Interference has been detected on several occasions, and captured on aerial video at the Palacios COM.

Generally, it is expected that suspended sediments will be greatest nearest the disturbance and decrease with distance. But delayed collections can result in lower levels near the disturbance and greater levels down current.

TSS Results from 10-1-22 Seagrass Disturbance Experiment:

Conditions: No clouds, 2m/s NE wind, current toward SW ~235 degrees; Later ~4.5 m/s wind


TSS Dry Weight (From Disturbance #1) [60 p-m]


TSS Dry Weight (From Disturbance #2) [60 p-m]

Total Suspended Solids Dry Weight collection from sediment plumes (2 Lines and 2 Disturbances) are consistent with distance in Disturbance 2 (both Lines) but showed surprising increasing trend with distance from the Disturbance Track for Disturbance 1.

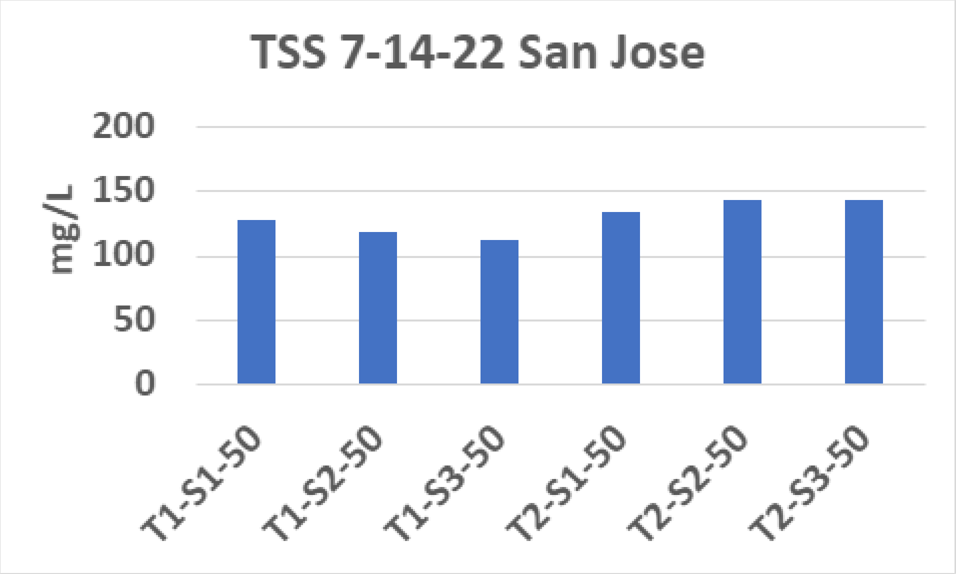
Note: UAS video shows TSS collection at Line 1 35 ft may have interfered with downstream Line 2 collections

Results are also consistent with clearing of sediment plume (settling and dispersion of suspended sediments) prior to TSS collection. UAS video at 24 min supports this.

Additional TSS measurements for San Jose Disturbance Experiments are presented below.

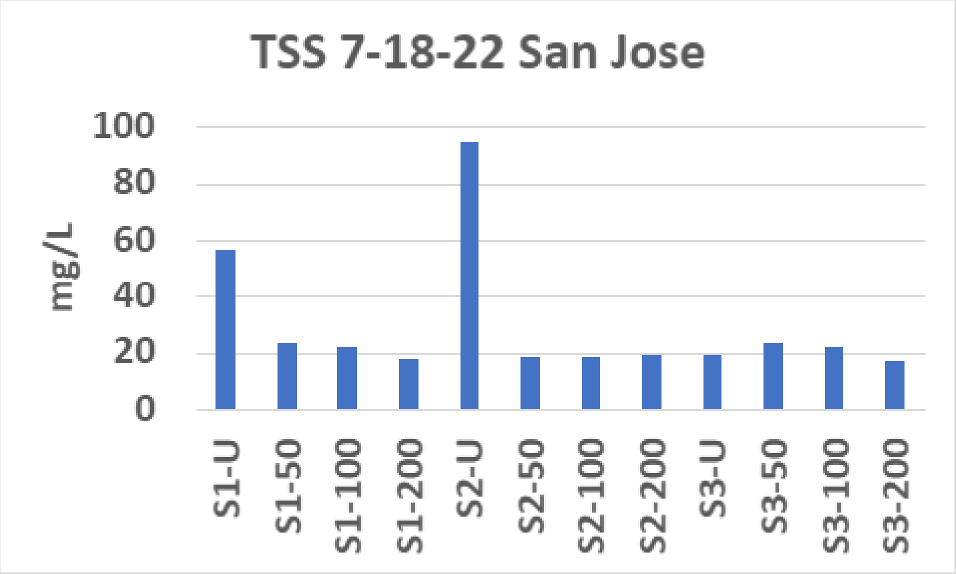
7-14-22 San Jose: In this experiment, the sediment plume never actually reaches the 50 ft pole. The lack of differences between tracks (treatments) is consistent with that observation.

Conditions: Very sunny, ~6m/s South/Southeast Wind. (T1 and T2 are two disturbances; S1 – S3 are different Tracks, and 5o represents the 50 ft measuring station)



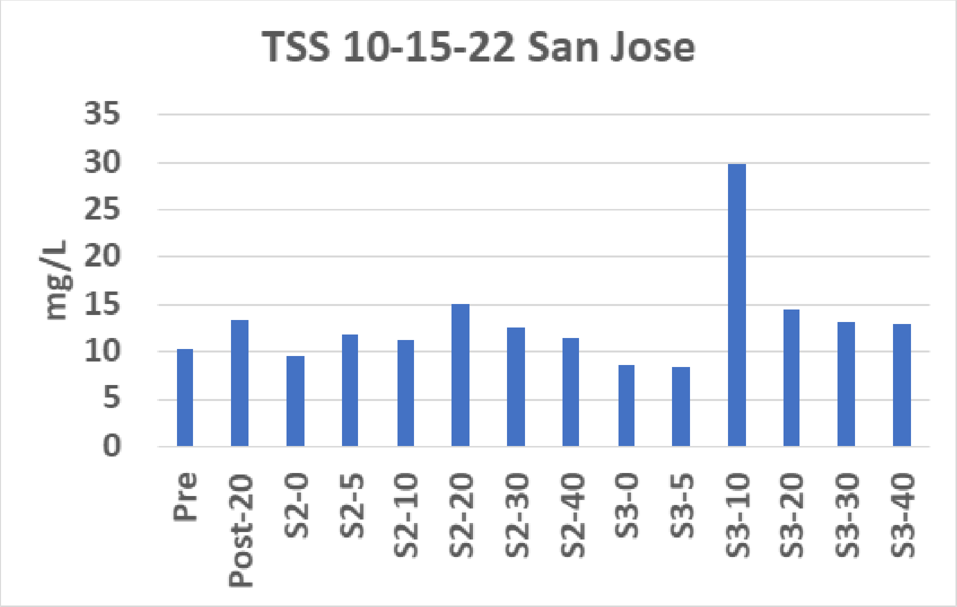
7-18-22 San Jose: In this experiment, the sediment plume moved backwards towards the Upstream poles and never reaches any downstream measuring stations. The high upstream values are consistent with this observation.

Conditions: Sunny, 12-15 mph SSE winds, ~5m/s 135 degrees wind. Low Water level.



10-15-22 San Jose: In this experiment, sediment plume TSS samples were collected at the 35 ft pole over time. Peak TSS sediment levels reached this position at approximately 10-20 minutes, and peak TSS levels from heavy disturbance Track 3 were twice those from the light disturbance in Track 2, indicating a proportionality of TSS levels with disturbance intensity.

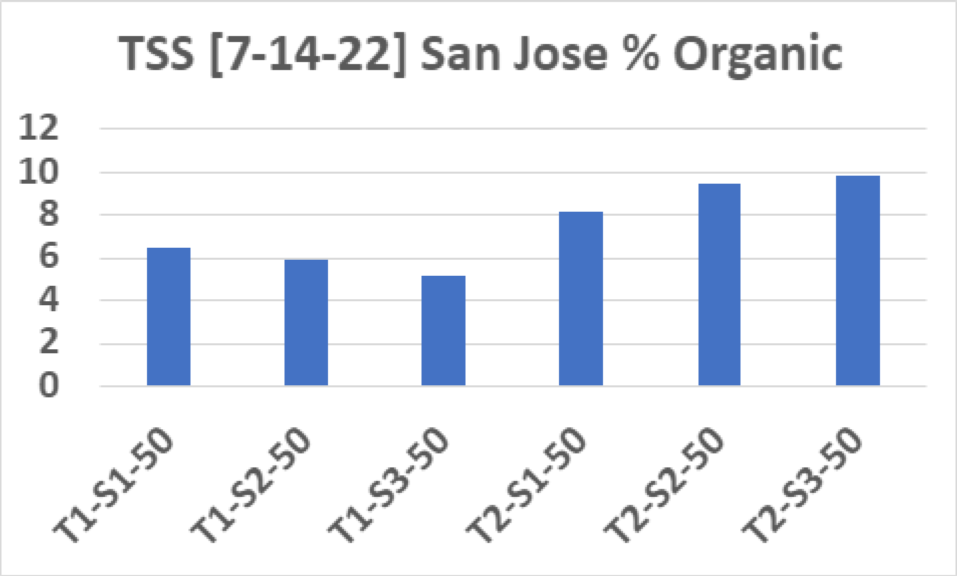
Conditions: Almost no clouds, ~3.6 m/s SE wind; Later ~3.3 m/s SSE Wind, Water was Clear in morning and later became turbid.



Samples collected at different **times** in disturbance tracks at 35 ft. TSS values peak at 10 or 20 min. Peak value at S3 (Heavy) is twice that at S2 (Light)

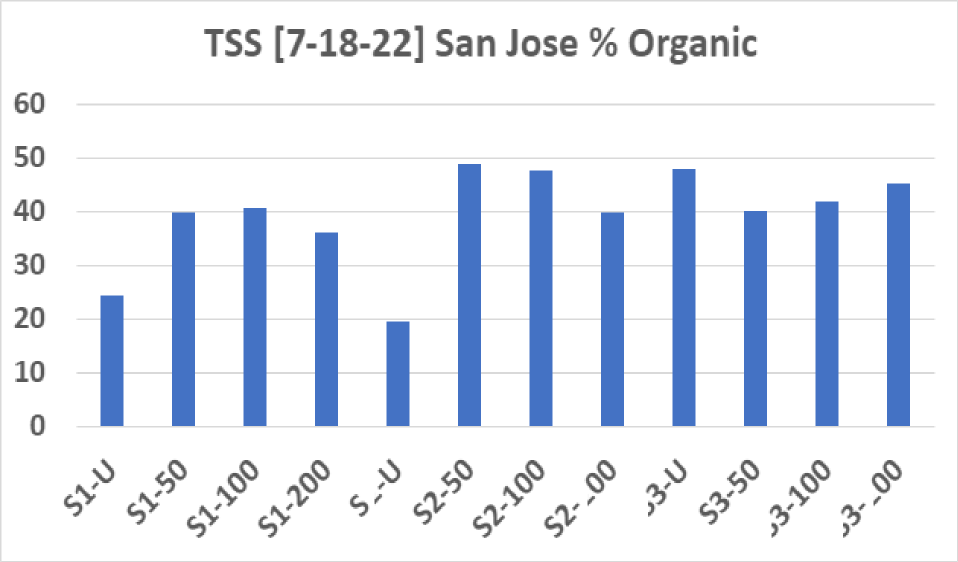
Percent Organic Component of TSS Samples (% OM)

7/14/22 Conditions: Very sunny, ~6m/s South/Southeast Wind.



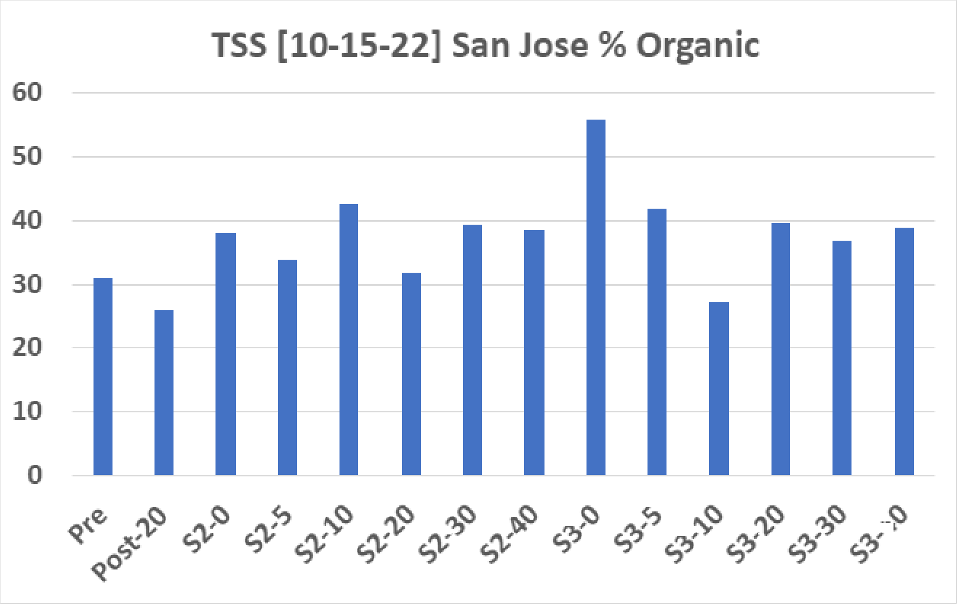
No clear trends in %OM between tracks., but higher values in Disturbance 2 (T2).

7/18/22 Conditions: Sunny, 12-15 mph SSE winds, ~5m/s 135 degrees wind. Low Water level.



No clear trends in %OM with distance, but Track 2 and Track 3 samples (including upstream control) were higher than for Track 1.

10/15/22 Conditions: Almost no clouds, ~3.6 SE Wind; Later ~3.3 SSE Wind, Water was Clear in morning and later became turbid.

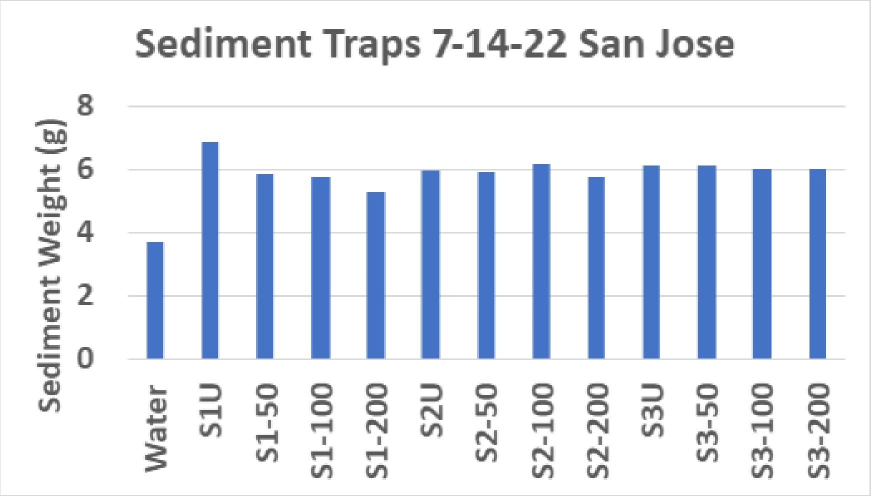


Samples collected at different **times** in disturbance tracks 2 and 3. No trends of % OM with time or track.

Settled Sediments (Sediment Traps)

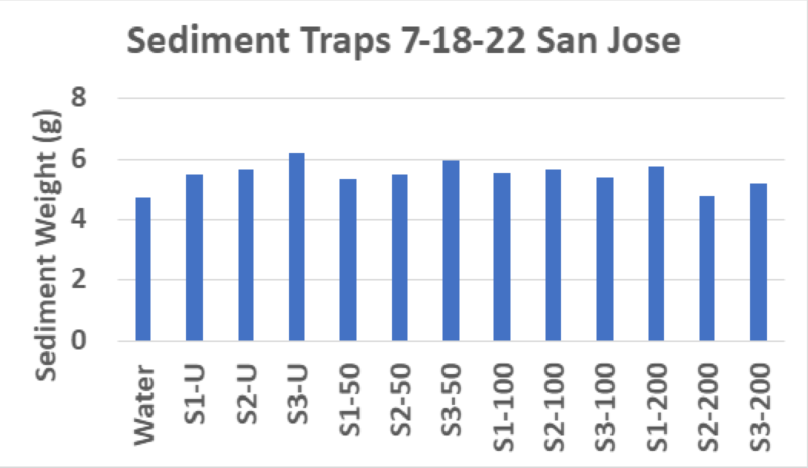
Sediment traps capture the readily settleable suspended solids. A gradient of decreasing sediment collection is expected with distance from the disturbance.

7/14/22 Conditions: Very sunny, ~6m/s South/Southeast Wind.



Other than a decreasing trend with distance in Control Track 1, no differences observed. Note that sediment plumes were not moving well this day.

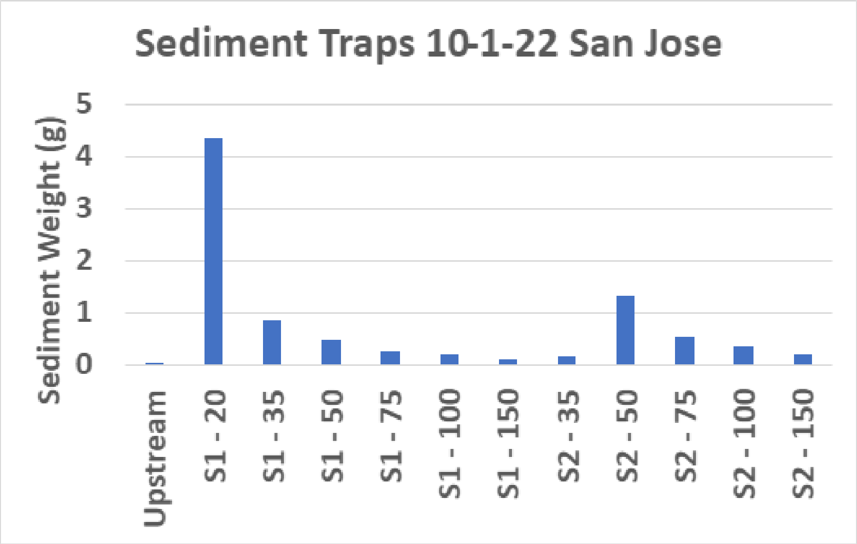
7/18/22 Conditions: Sunny, 12-15 mph SSE winds, ~5m/s 135 degrees wind. Low Water level.



For Ups and 50 ft collections slight increase from Track 1 to Track 3. No other differences. Note that sediment plumes were not moving well this day.

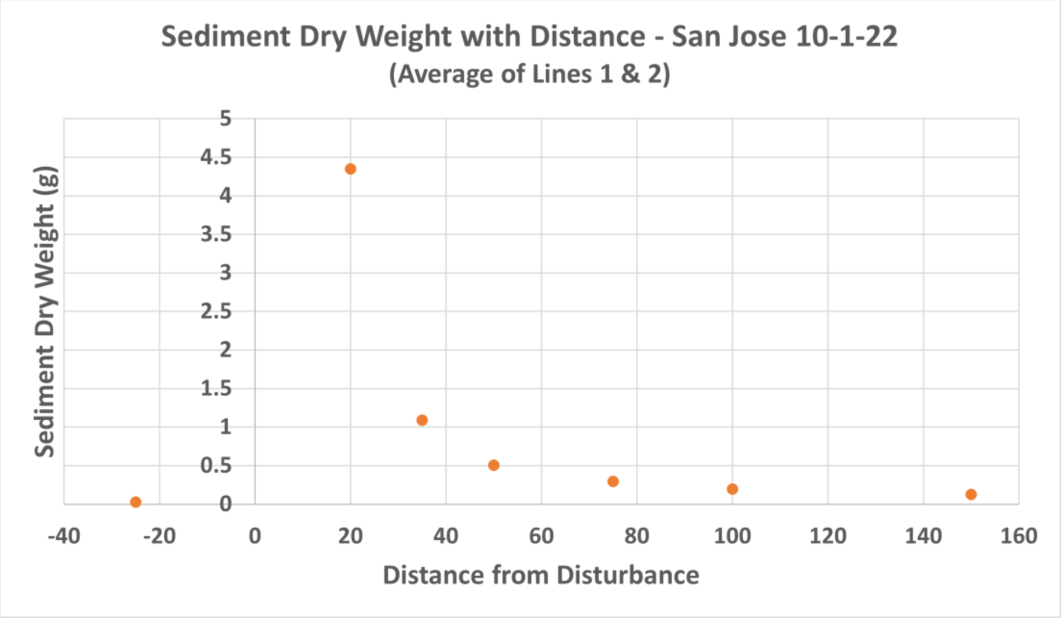
10/1/22 Conditions: No Clouds, NE to ENE 10mph Wind. ~1.7 m/s NE Wind; Later ~4.5 m/s

For this experiment, S1 and S2 represent the two lines of measuring stations setup in dual fan array with a perpendicular orientation to the Tracks.



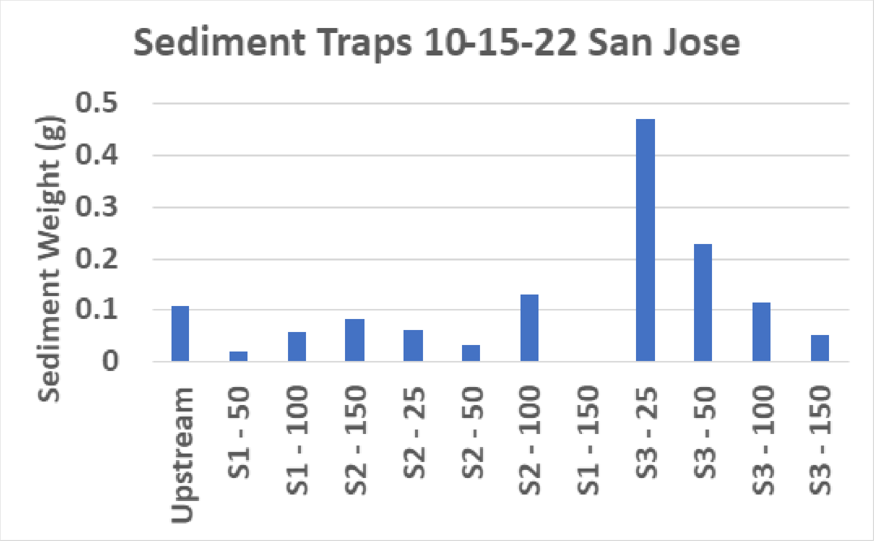
Observe decreasing trend in sediment trap collection with distance from the disturbance. All downstream collections are greater than for the upstream control. Note that Line 2 trap collection had a peak value at 50 ft rather than at 35 ft (no 20 ft for this line). This could be due to alignment or a disturbance during collection.

Plot of average sediment collection (lines 1 & 2) with distance on scaled X-axis (Distance) for two 60 p-m disturbances



Most settleable solids from the disturbances are settled within the first 50 ft, but values above that of the upstream control persist to at least 150 ft. Recall that sediment plume was visible out to at least 300 ft.

10/15/22 Conditions: Almost no clouds, ~3.6 m/s SE wind; Later ~3.3 m/s SSE Wind, Water was Clear in morning and later became turbid.

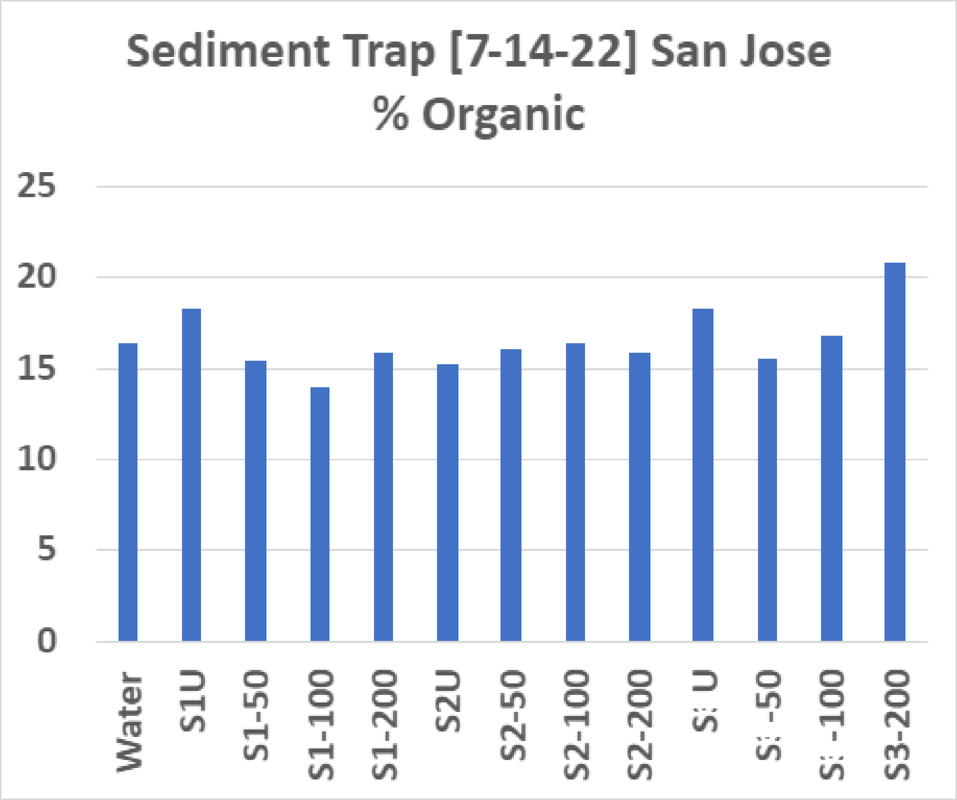


Track 3 Heavy disturbance shows decreasing trend of sediment collection with distance.

% Organic Component (Sediment Traps)

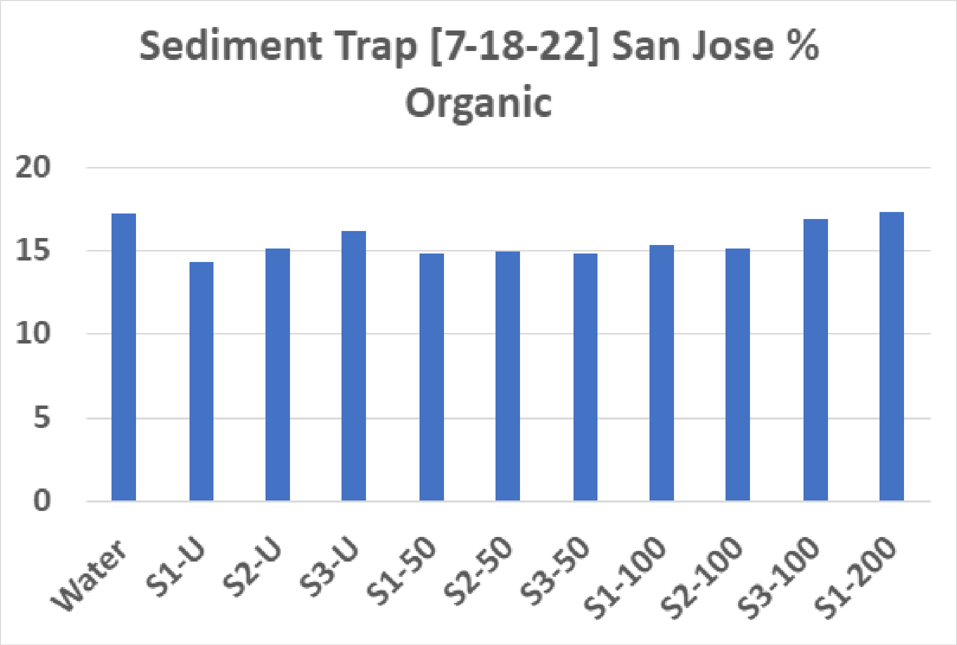
As for TSS, it is expected that in sandy sediments, settled particulates will exhibit low % OM, whereas material derived from non-calcareous biota will have a relatively high % OM. Also, it is predicted that suspended sandy sediments will settle quickly, or near the disturbance, while organic particles are likely to stay suspended longer and travel farther. So % OM provides an important characterization of the settled solids. Most % OM values observed for seagrass sediment plumes ranged between 5% - 20% OM, with a few values spiking very high.

7/14/22 Conditions: Very sunny, ~6m/s South/Southeast Wind.



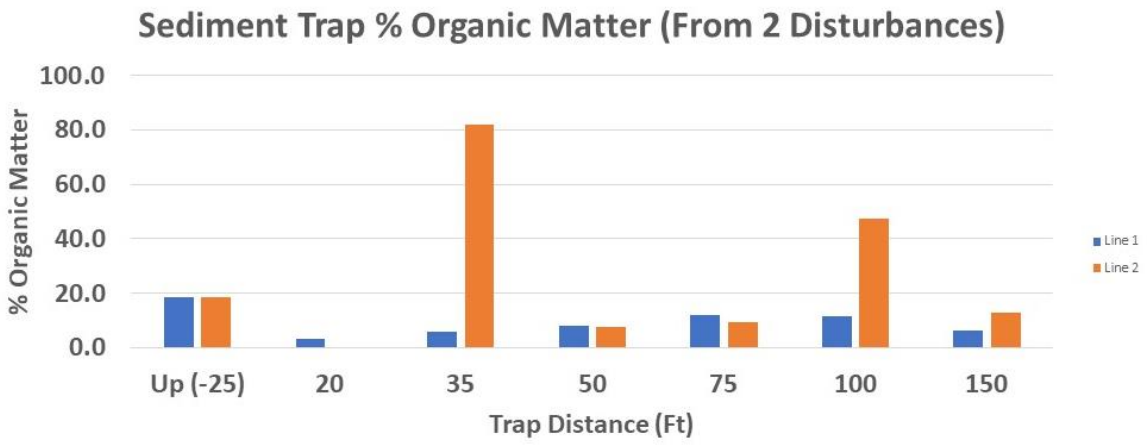
Increasing %OM with distance only in Track 3, but the Up value was within that range, so no discernable trend.

7/18/22 Conditions: Sunny, 12-15 mph SSE winds, ~5m/s 135 degrees wind. Low Water level.



No observable trends for % OM between tracks or with distance. There was little sediment plume movement with disturbances.

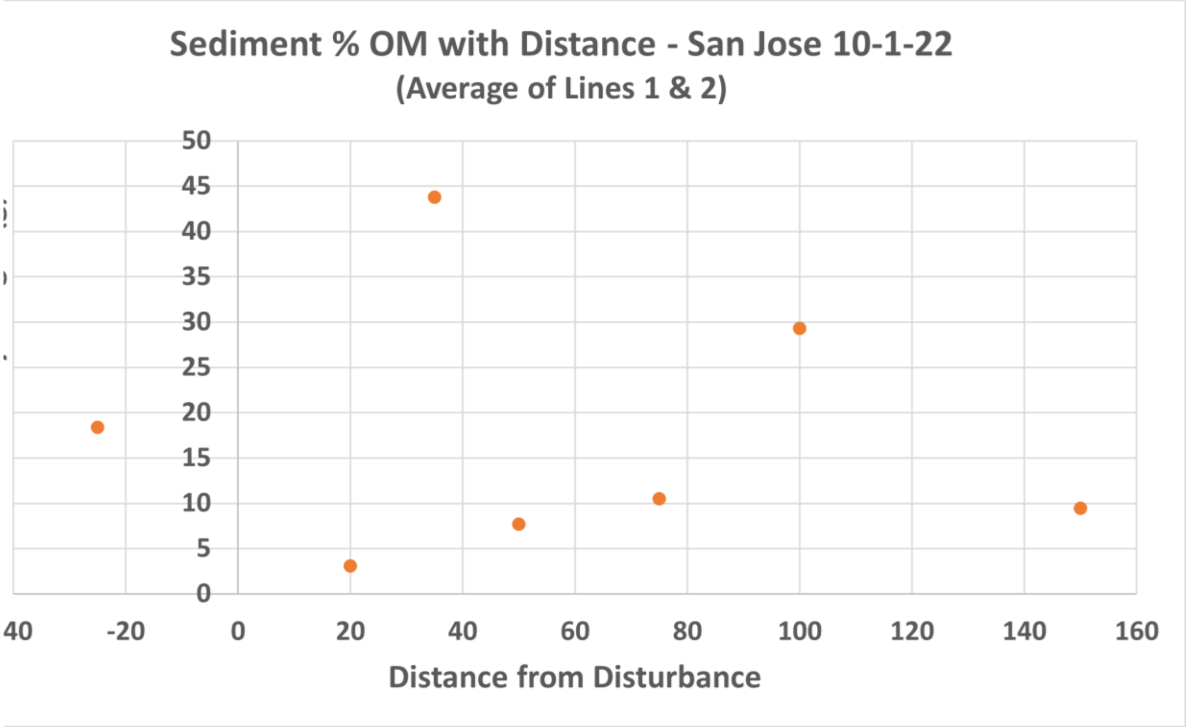
10/1/22 Conditions: No Clouds, NE to ENE 10mph Wind. ~1.7 m/s NE Wind; Later ~4.5 m/s (No 20 ft pole for Line 2)



Data consistent with an increasing % OM with distance indicative pf preferential settling of non-organic sediments with distance. Values at 35 ft and 100 ft for Line 2 may indicate interference during sample collection. Alternatively, there could have been a piece of detritus or an invertebrate that entered the sediment trap, but we filter out anything larger than 1 mm in size.

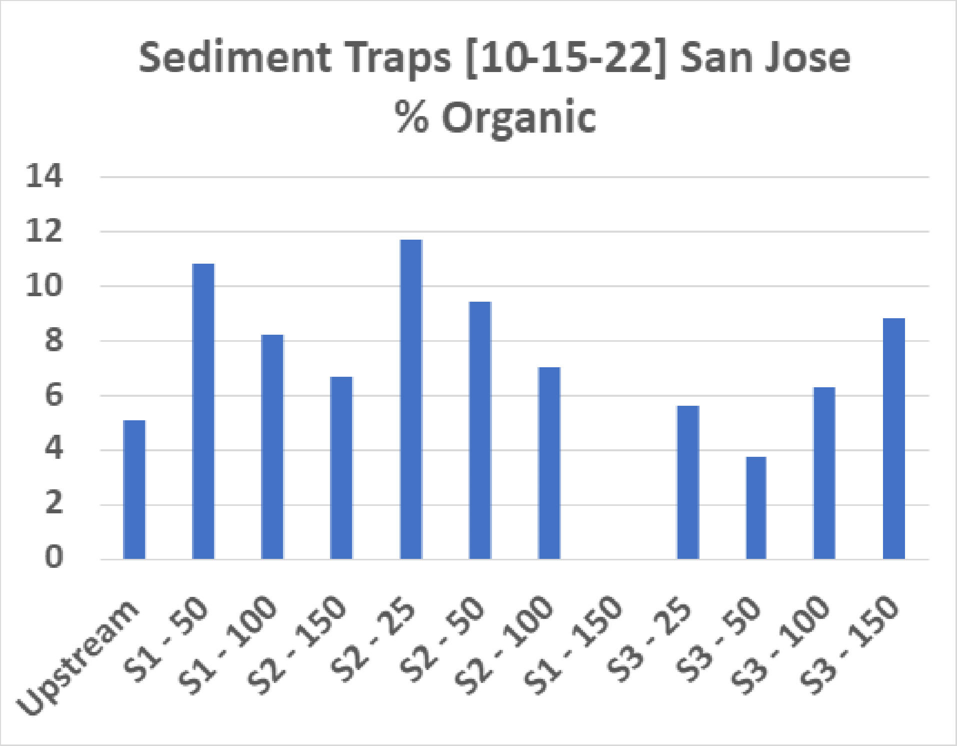
Similar data were observed from both lines of measuring stations. Note that sediment collections decreased with distance from the disturbance. At 150 ft most of the suspended sediments (by weight) have settled or dispersed. Despite this, sediment plumes are visible out to 300 ft. These probably comprise relatively light weight, low density and or fine-sized suspended particles that do not settle readily. Note that the % OM at 150 ft was less than that at the Upstream station (~5%-10% at 150 ft vs ~20% at Up). This would suggest that there are still inorganic suspended particles being carried in the sediment plume at this distance.

Plot of average % OM from sediments (lines 1 & 2) vs distance on scaled X-axis (Distance) for two 60 p-m disturbances



However, this data does not provide evidence of relationship between % OM and distance (a best fit linear trendline of downstream data points is nearly horizontal; not shown).

10/15/22 Conditions: Almost no clouds, ~3.6 m/s SE wind; Later ~3.3 m/s SSE Wind, Water was Clear in morning then became turbid.



Tracks S1 and S2 show decreasing % OM with distance, whereas Track 3 (Heavy) shows a generally increasing trend with distance.

Results are mixed concerning a relationship between % OM and distance from disturbance. Data from the different individual experiments are analyzed collectively at the end of results to further explore TSS, sediment trap, and % OM relationships with distance from disturbance.

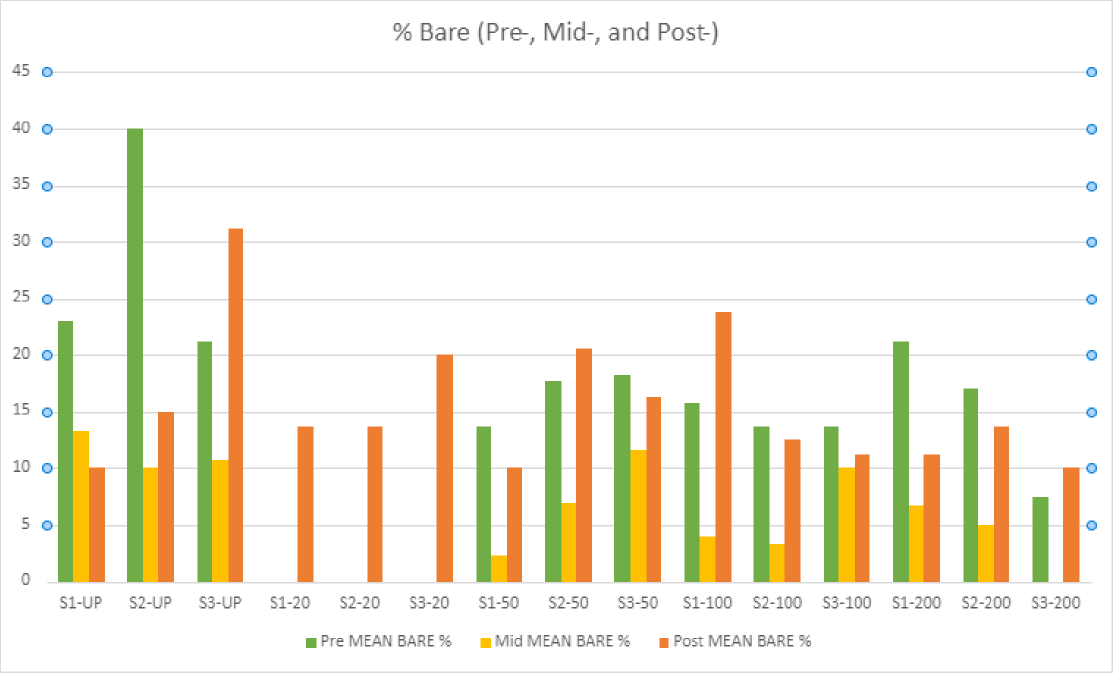
Seagrass Bed Biological Measures

Different parts of the San Jose seagrass study site received a cumulative total of 811 p-m (Tr 3), 455 p-m (Tr 2) or less (Tr 1) of simulated sediment disturbance over the course of this project. A variety of seagrass biotic indicators were measured pre-, mid-, and post-disturbance experiments to determine if these levels of sediment plume impact had any effects on the status or functioning of the seagrass system. The pre, mid- and post- sampling timepoints are also referred to as "milestone" sampling timepoints. In some cases, seagrass samplings were performed at other times *in addition to* the milestone timepoints.

Seagrass Bed Indicators

Indicator metrics of seagrass bed quality include % cover by seagrass, leaf density, root to shoot biomass ratio and canopy height (Neckles et al. 2012; McMahon et al. 2013). Invertebrate sampling for abundance and diversity metrics was also performed to provide an indicator of seagrass bed function. Sampling for these measures took place in each zone of each track, nearby to where the measuring stations were located in the original experimental design. Sampling occurred Pre-experiment, at one or more times during the experiment, and then again Post-experiment.

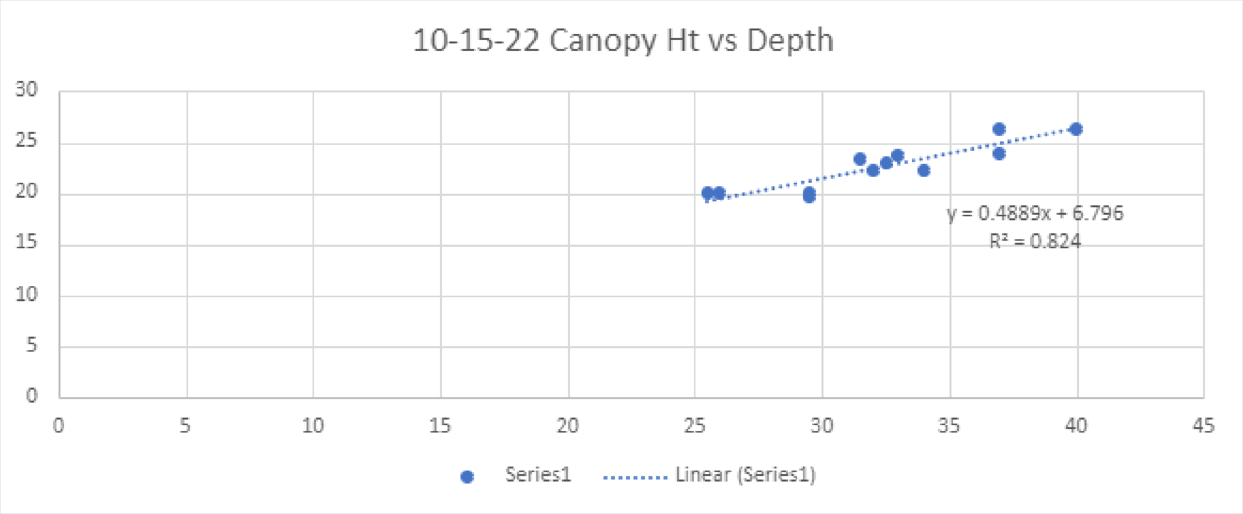
Seagrass Cover Quadrats

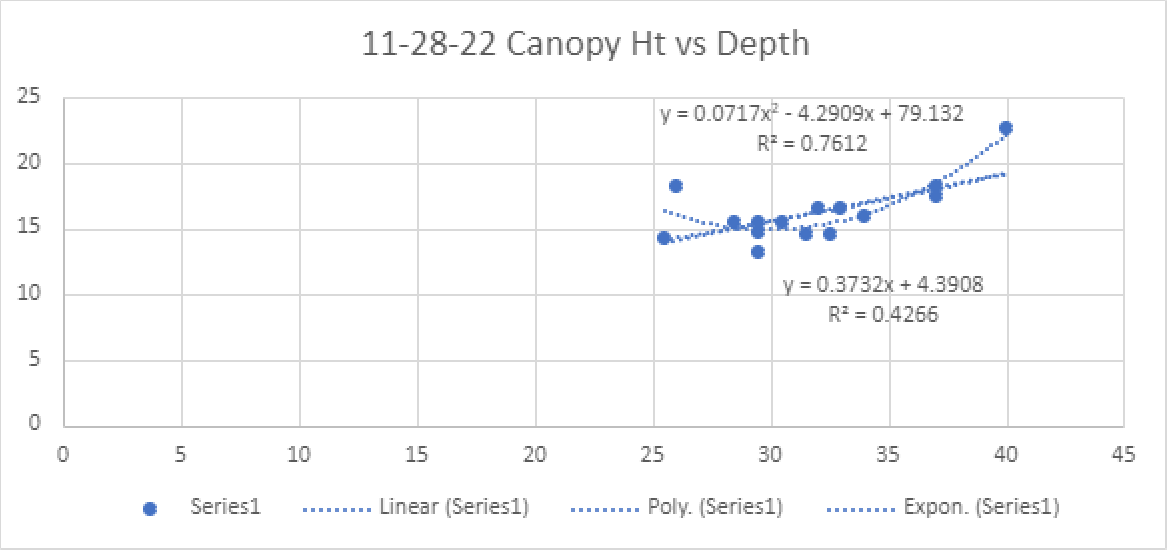


This plot shows the mean (n = 4) of % Bare for haphazardly located quadrats within each zone of each track at the San Jose seagrass study site, measured Pre-, Mid- and Post- experiment. No significant differences were detected in comparisons of Pre vs Post among tracks or distances. This suggests that the sediment plume treatments did not affect % seagrass cover in this short term experiment.

Canopy Height to Depth Correlation

Seagrass canopy height was measured during quadrat surveys. Results from canopy height were plotted against a depth gradient using the depths of different locations along the track. In two separate sampling dates, canopy height was correlated with depth, achieving R squared values of .42 and .82 respectively. Seagrass blade length data is presented later to illustrate the seasonality of this metric.

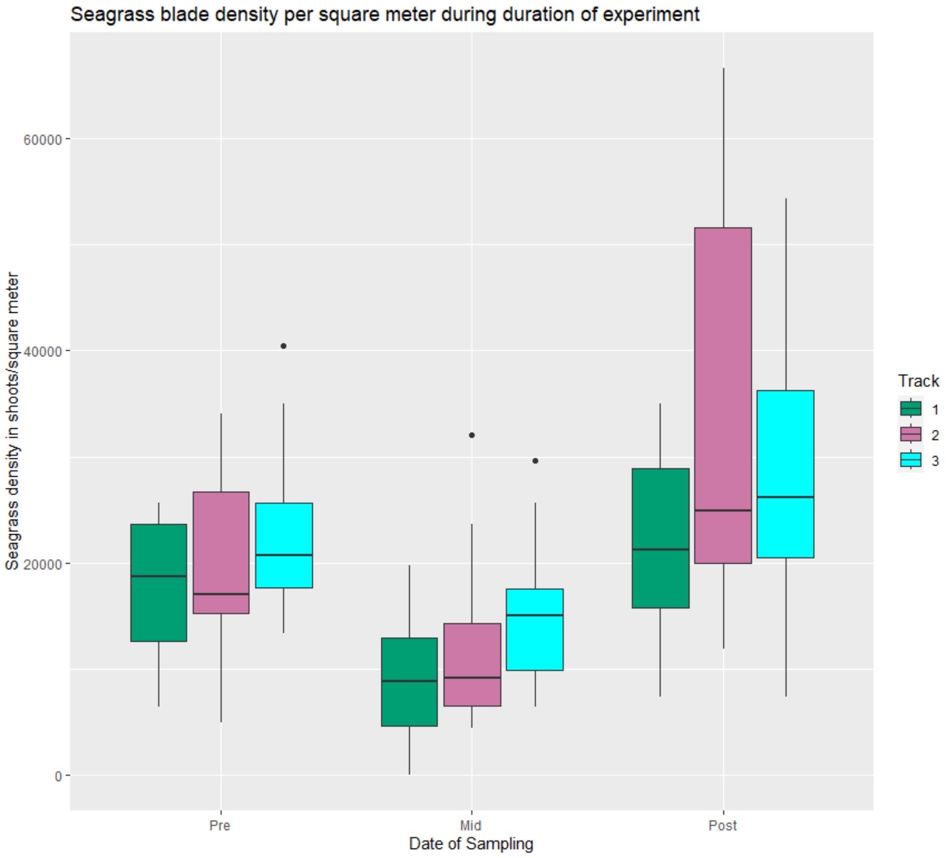




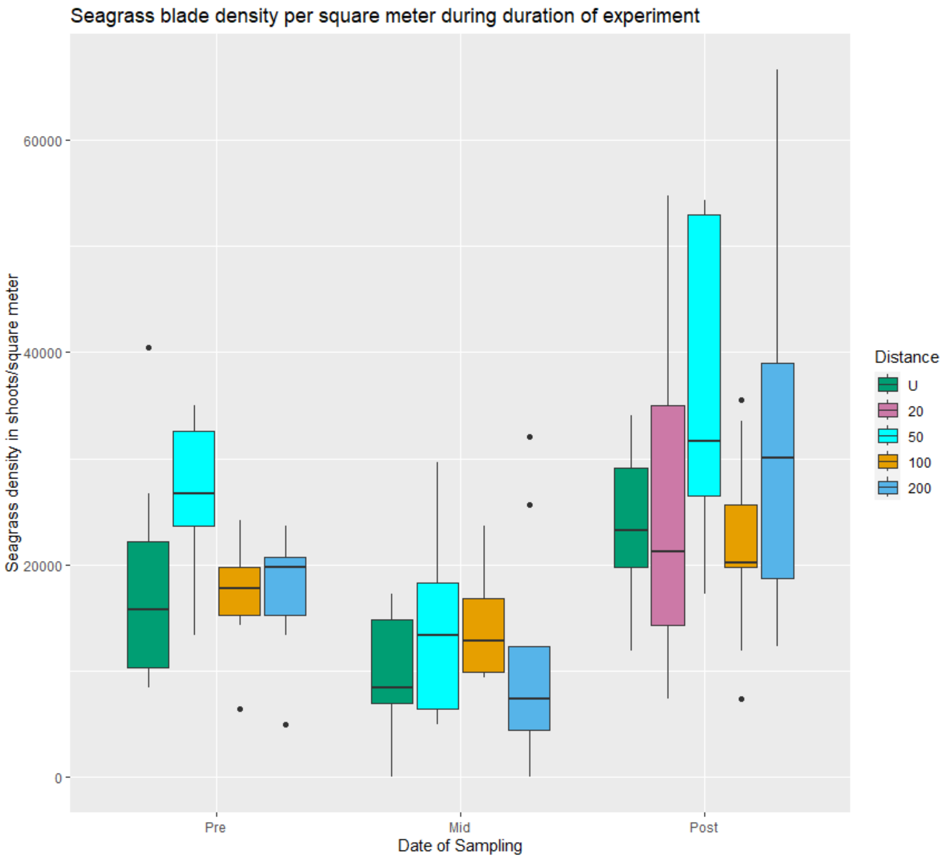
11-28-22 Canopy Ht vs Depth
y = 0.0717x² - 4.2909x + 79.132
R² = 0.7612
y = 0.3732x + 4.3908
R² = 0.4266
25
20
15
10
5
0
0
5
10
15
20
25
30
35
40
45
Series1
Linear (Series1)
Poly. (Series1)
Expon. (Series1)

Leaf Density and Biomass

Seagrass leaf density and biomass were determined from seagrasses harvested using the ring collection method (all shoots within a PVC ring of 0.00203 m^2; 3 haphazardly placed rings per sampling site).



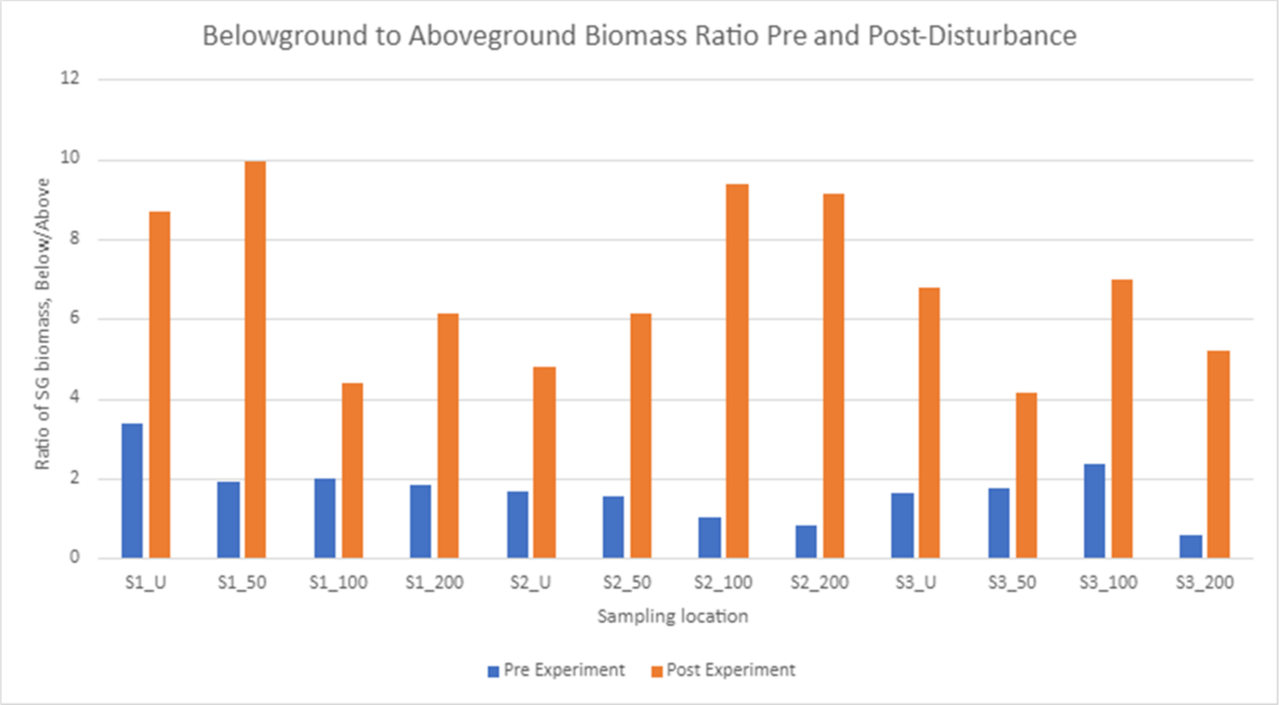
Leaf densities were not significantly different compared between treatment tracks or across different sampling times.



Seagrass leaf density compared across sampling times for different distances from the disturbance baseline. All three treatment tracks are combined here. There were no significant differences with distance or sampling time. A similar pattern was seen comparing across the distance gradient, but greater levels of variation were observed in the post experiment sampling of seagrass density.

Root to Shoot Biomass Ratio

The Root to Shoot Biomass Ratio is an important indicator for seagrass beds. Because it necessitates destructive core samples, this was only performed pre- and post- experiment. Presented values are the mean of 3 core samples from each sampling location.



The mass Ratio of Belowground seagrass material (Roots, Rhizomes) to Above ground seagrass material (Leaf, Sheaths, Epiphytes) is compared Pre- and Post- disturbance experiments. The blue bars represent pre-experiment sampling (June 2022) and the orange bars represent post-sampling (December 2022). The Post experiment samples had an average of ~5 times the amount of belowground material to above ground, compared to pre-sampling where the ratio was closer to 1.

Sampling in June was done in conjunction with the collection of invertebrates using 10 cm diameter core samples which were screened through a fine mesh and seagrass was then removed. In December sampling was done independently from invertebrate cores using a similar method of coring, screening and sorting the seagrass shoots, roots, and rhizomes. There were no significant differences in any comparisons between treatment tracks, along distance gradient within tracks or between the 50 ft zone A area of each track.

Invertebrate Indicators

Abundance

The overall mean (±1 S.D.) of invertebrate densities per m^2 was 1250 (664) including all sampling periods, tracks, and times. The most abundant invertebrates (with mean densities per m^2) were: the small gastropod *Nassarius* sp (309), the polychaete *Capitella capitata* (203), another Capitellid polychaete, *Mediomastus* cf *ambiseta* (103), and the small bivalve *Eurytellina alternata* (85). The nereid polychate (*Laeonereis culveri*, 24 specimens) and other unidentified nereids (sometimes missing key parts, 33 individuals) were also relatively abundant. The most common crustacean was the grass shrimp *Palaemonetes pugio* (18 individuals). Cumulative species by samples is shown in Fig. 1.

Initial assessment for testing hypotheses indicated that the Upstream site was not really a good “control” or “reference” for invertebrates in this study. Sediment appeared firmer, the water was a bit shallower, and possibly other small differences that might not make a difference for seagrass per se or epiphytes, but did cause a severe restriction in macrofauna. Thus, this site was dropped from analysis and is not discussed further here.

Total invertebrate abundance was tested as a function of treatment distance downstream from disturbance and trip (Fig. 2), however analyses showed that trip (sampling time) did not have a significant effect on abundance but did inflate AIC and this variable was dropped from the final model. Additional tests indicated that track (different transects with different levels of disturbance) did not have a significant influence on invertebrates. The final model tested against the negative binomial distribution which provided a much better fit than the Poisson. which showed a significantly greater densities nearer the disturbance source (response and parameter estimates in natural log units, model: **ln(total.abundance) = 2.5878 – 0.004 (dist. In feet from disturbance); p < 0.0039).** Number of species was tested against the Poisson distribution, and did not vary significantly with either distance from disturbance (or trip (sampling time) or track (transect).

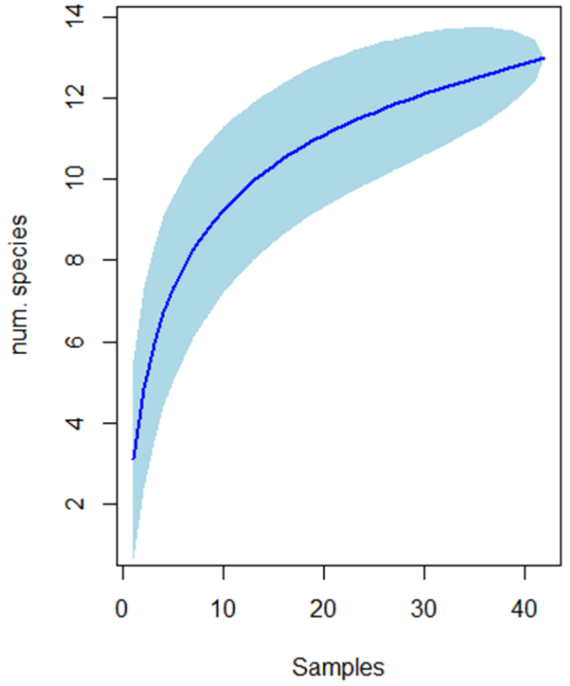


Fig. 1. The cumulative number of species recorded vs samples collected.

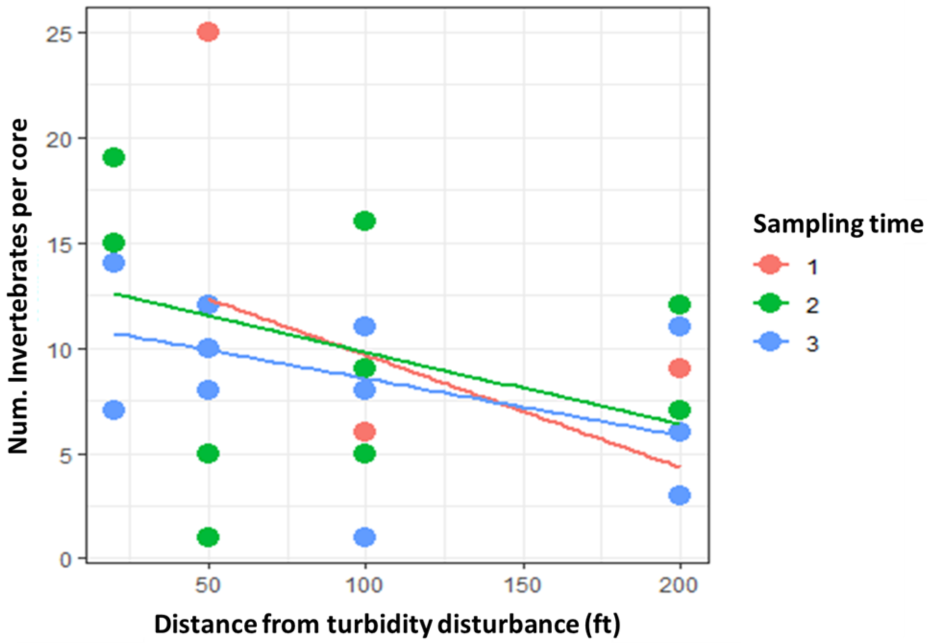


Figure 2. linear trend of decreasing invertebrate abundance with increasing distance from turbidity source for the 3 sampling times. Note sampling occurred 1 week following each disturbance.

Diversity and community dissimilarities

Mean species diversity (the inverse of the Simpson's Index) was 0.465 (0.227 S.D.), a moderate level of diversity. Diversity did not vary substantially among distance treatments (distance from disturbance, ($t_{5,27}$= 1.451, p= 0.238)) or tracks or time or their interactions.

Community analysis was first visualized as NMDS ordination using the metaMDS function in the library vegan. The ordination of species dissimilarities and fit plot are shown in Fig. 3.

PERMANOVA using adonis2() function in the vegan library indicated a marginally significant variation in of community dissimilarity at different distances from disturbance ($F_{1,41}$ = 2.12, P < 0.061) but not differ by track ($F_{2,41}$ = 1.49, P < 0.151) or time ($F_{2,41}$ = 0.819, p = .586). The interaction term was not significant ($F_{2,41}$ = 0.52, P =.878). Thus, there was support for the hypothesis that disturbance affected community composition but not species richness or the inverse of Simpson's diversity index. The increasing dissimilarity with increasing distance from turbidity source correlates with the increasing abundance recorded with increasing distance. This may be indicative of additional species being attracted to the disturbance as stirring of the benthic sediments may release food sources for detritivores and those that depend upon them.

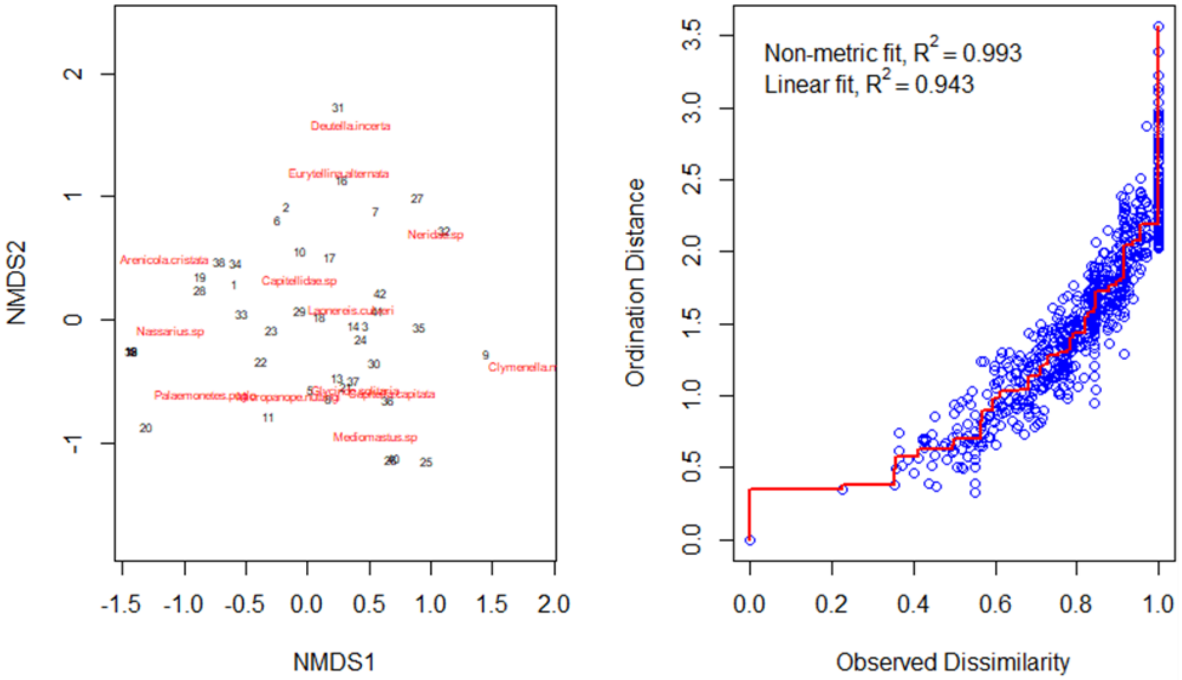


Fig. 3. NMDS (left) of the invertebrates, and Shepard stress plot (right) showing a high goodness of fit of the ordination.

Seagrass Plant Indicators

The seagrass plant indicators provide insight into the status and functioning of the seagrass plants. Indicator measures include leaf length, leaf biomass, and metrics of epiphyte accumulation. Two different seagrass sampling strategies (described below) were used for equity in representation of different plant sizes and to facilitate data aggregation and comparisons in different ways.

The experimental design of the seagrass experiment facilitated comparisons between treatment Tracks, along different distances from the disturbance source, and across different sampling times (both seasonal and cumulative experimental effects) to determine if the simulated sediment plumes are impacting the seagrasses.

Sampling by Rings

Sampling by the collection of all seagrass shoots within a 5.08 cm (2-inch) diameter ring (0.00203 m^2) was performed during the Months of October and November to observe differences with progress of the sediment disturbance experiment and across seasons as well. Collection of whole shoots may bias the samples towards larger shoots, whereas this "Ring Collection" would include representation from younger, shorter shoots. Furthermore, the data can be used to calculate a shoot density per seagrass bed area (presented above), albeit only over a very small area subsampled.

Sampling by Shoots

Sampling by the collection of 15 whole seagrass shoots was performed Pre-, Mid- and Post-Disturbance Experiment (Months June, September and December, respectively) to observe differences with progress of the sediment disturbance experiment and across seasons as well.

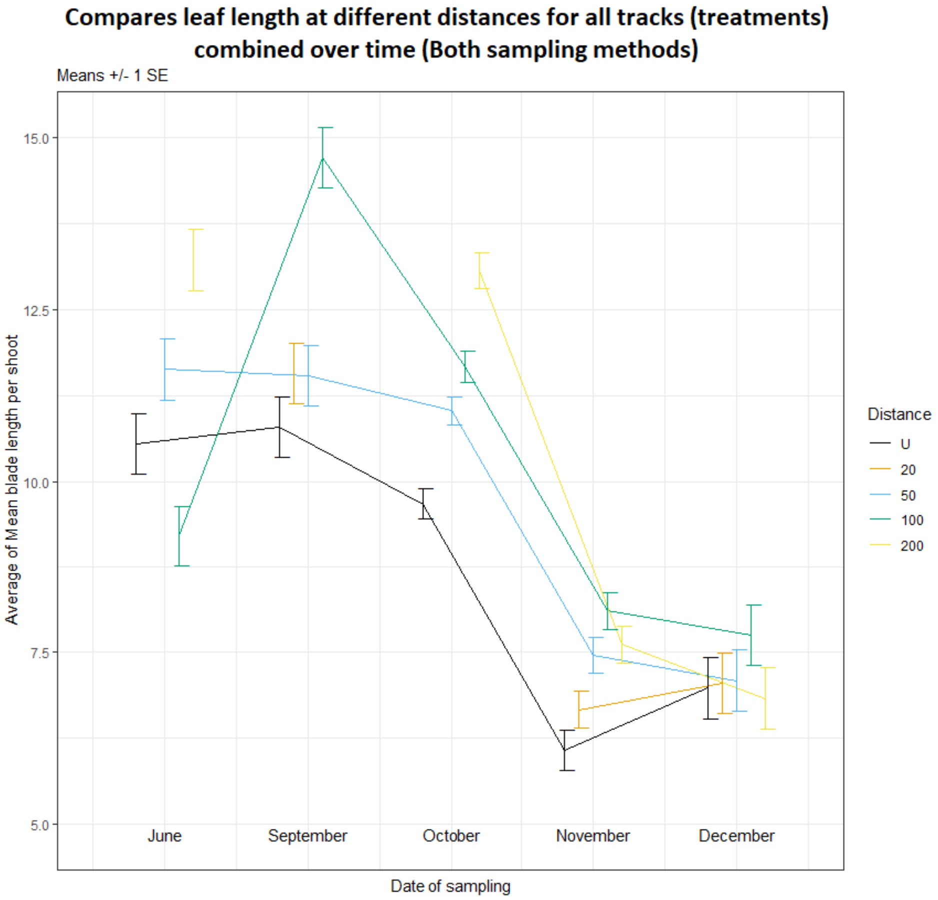
All Data Compared on a Per Shoot Basis

Alternatively, seagrass samples acquired by both sampling methods can be collectively analyzed on a mean per-shoot basis. For ring samples, the sample sizes will vary somewhat based on the number of shoots in the sampled ring. This ranged from 22 to 95 shoots per ring (= 0.00203 m^2).

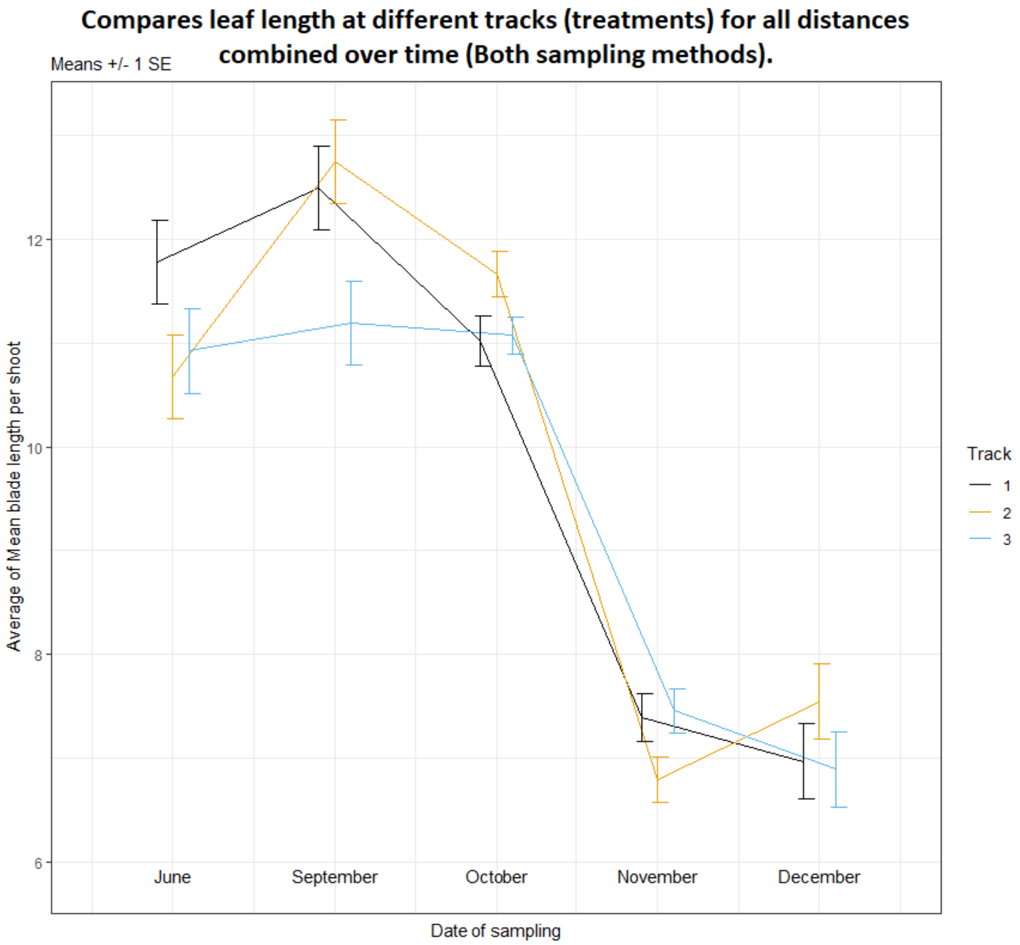
Data for the 15 whole shoots sampling method are presented for milestone sampling time points (Pre-, Mid-, and Post Disturbance). More frequent monthly sampling analyses are presented on a per shoot basis as represented by both sampling methods combined.

Morphology: Leaf Length

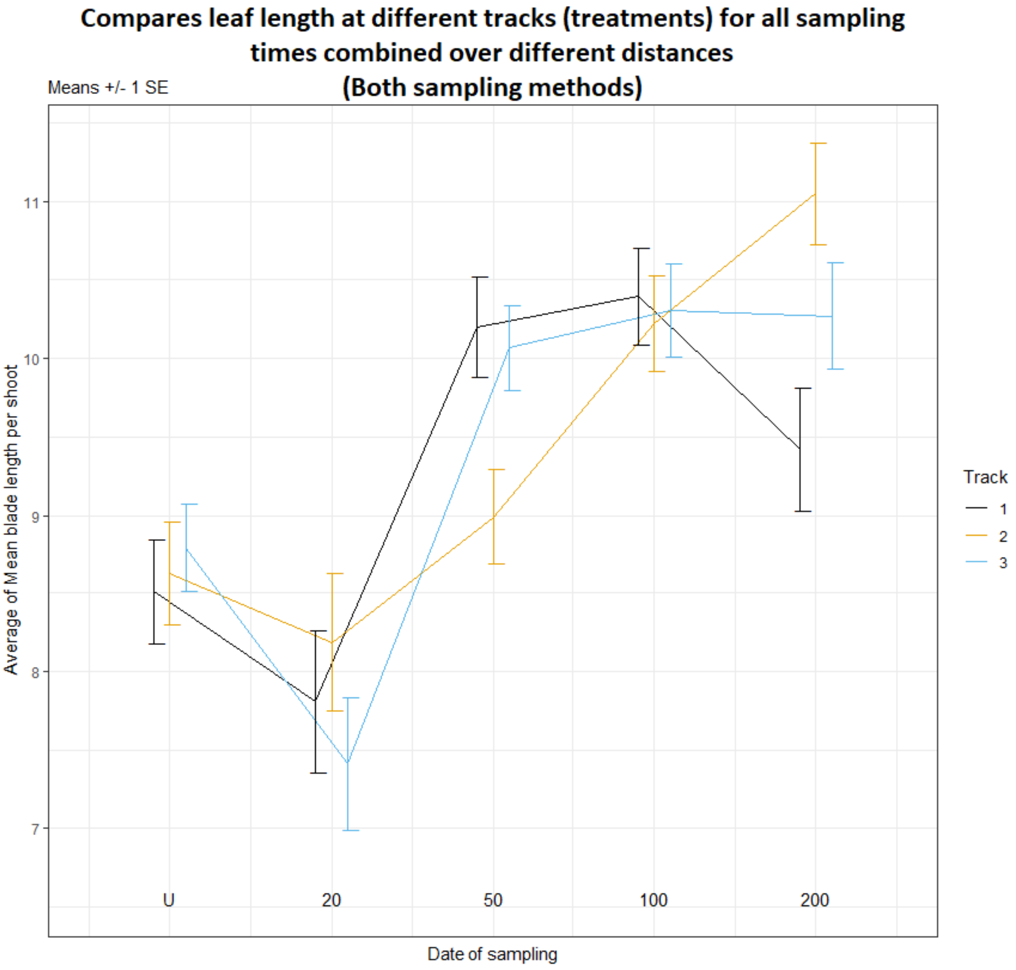
Leaf lengths of samples from each measuring station were compared between distance from disturbance, treatment tracks and sampling times.



Seagrass blade length exhibited the expected seasonal variation (decreasing from late summer highs into late autumn). On top of that variation, there were differences between upstream and different distances from disturbance when all treatment track samples were aggregated. This difference (increasing length with distance from Up or disturbance) correlates with increasing depth as was shown above. The Up locations on average were about 10 cm shallower than at the 200-foot markers.



Comparison of the seagrass blade length between treatment tracks did not reveal any significant differences across sampling times when all distances were aggregated, except for September sampling where track 3 mean blade length was lower than the other two.



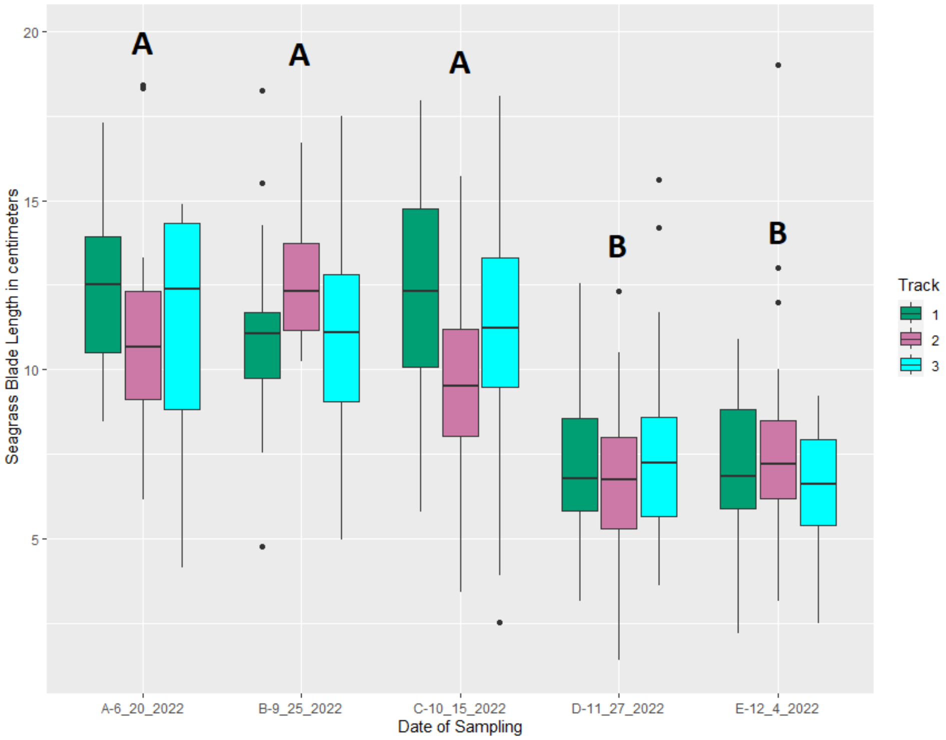
Comparison of the seagrass blade length between treatment tracks with a two-way ANOVA did not reveal any significant differences between tracks across distances when all sampling times were aggregated.

Overall, there was a non-significant decrease in blade length at 20 ft down-current compared to Up and 50 ft down-current. As depth increased at the 50-, 100-, and 200-foot markers, mean blade length increased, but the differences between tracks were non-significant.

Focused Comparisons of Samples from 50-ft Zone “A”

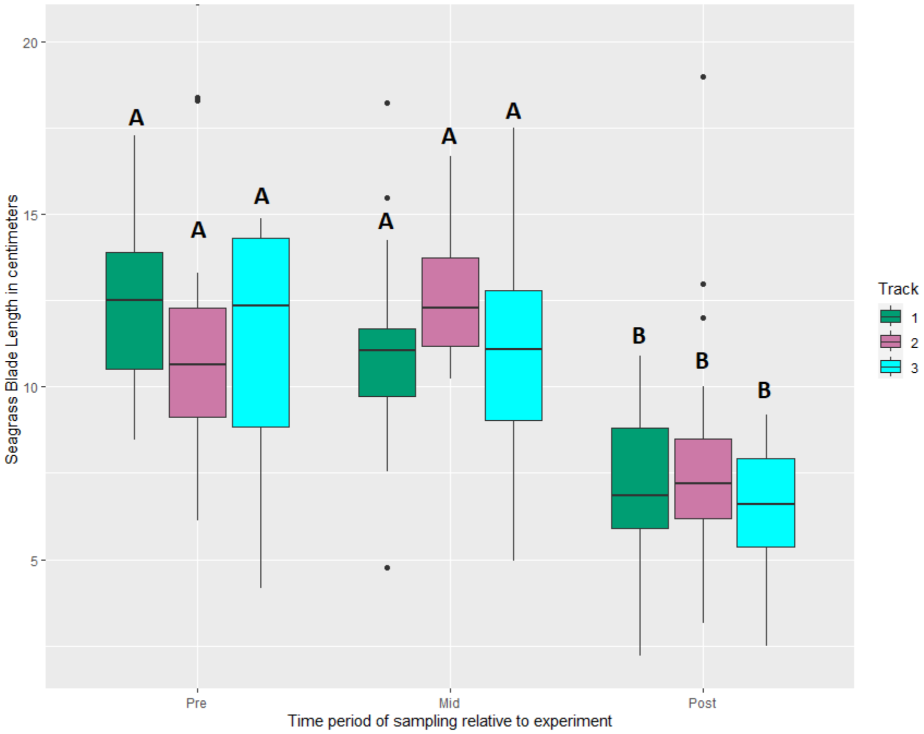
As our objective was to compare seagrass indicators between sites with different levels of sediment disturbance. The seagrass samples exposed to the greatest sediment disturbance are located in the 50 ft area (Zone “A”) of Track 3 (Heavy Disturbance). The most comparable site with the least sediment disturbance would be the 50 ft area (Zone “A”) of Track 1 (No Disturbance). Intermediate results were predicted for a light disturbance (Zone “A” 50 ft area of Track 2). Thus, the comparison *most* likely to reveal a difference (or conversely refute the hypothesis that the sediment disturbance impacted the seagrass) would be a comparison of seagrasses at the 50 ft distance across Tracks 1-3.

2-way: Compares Track (treatment) and sampling time for samples from the 50-ft Zone "A" only.



Seagrass length varied significantly with sampling time (season) but not between tracks within the first 50 feet (zone "A") of the experimental area. This comparison presents all seagrass samplings (from both methods) together.

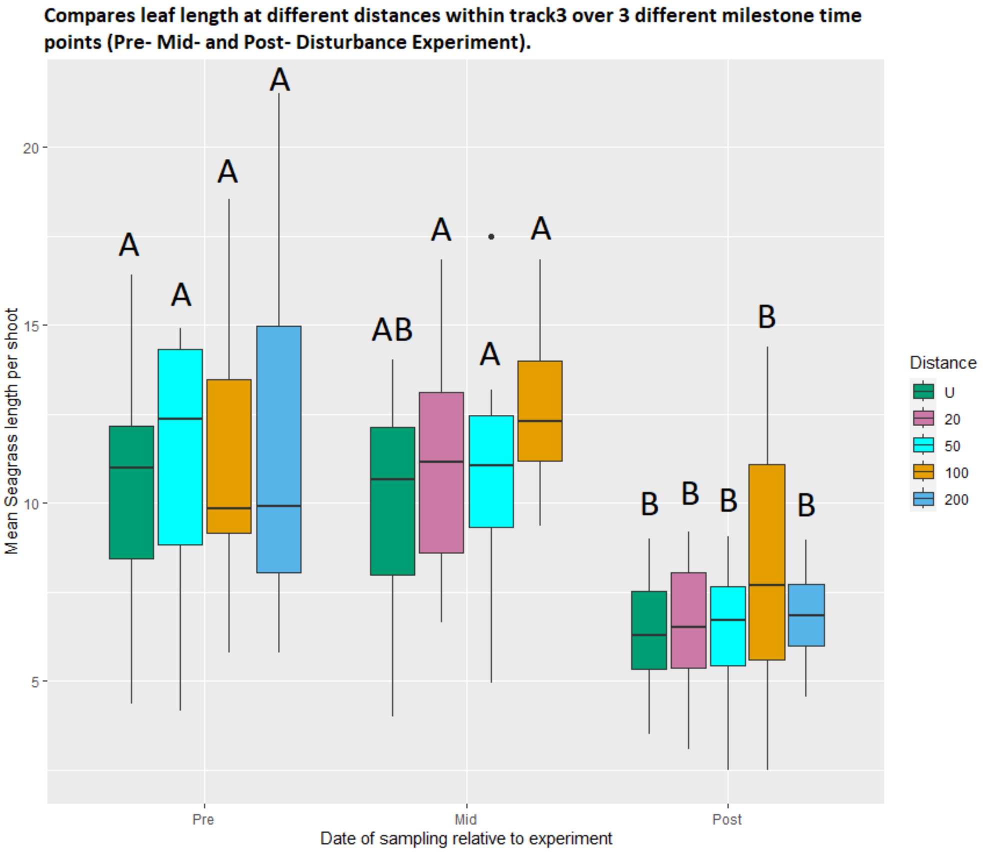
2-way: Compares Track (treatment) and milestone sampling times for 15 whole shoot samples from the 50-ft Zone "A".



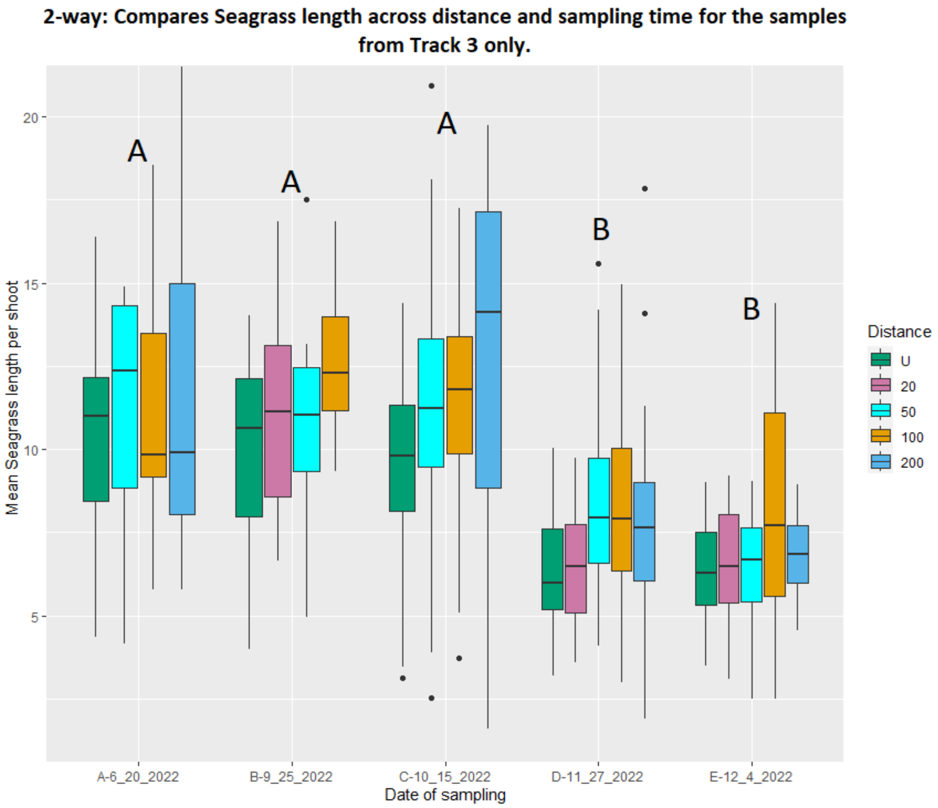
This comparison of mean seagrass blade length across tracks within the first 50 feet (zone "A") shows data for only the 15 shoots collection method used at the milestone time points. Again, there were no significant differences between treatment tracks.

Focused Comparisons of Samples from Track 3 (Heavy Disturbance) with Distance From Disturbance

An alternative focused comparison for samples from the 50 ft heavy disturbance area (Track 3 50 ft) is along the same track across different distances from the disturbance source. Sediment plune intensity is expected to diminish with distance and thus the potential impacts to seagrass should diminish similarly. The distance gradient also corresponds to a depth gradient from ~45cm to ~55 cm.



Comparison of seagrass length within track 3 across the distance gradient and time. ANOVA markers show no significant differences between distances from disturbance within Track 3 (Heavy) across the milestone sampling times which exhibit the seasonal variation.



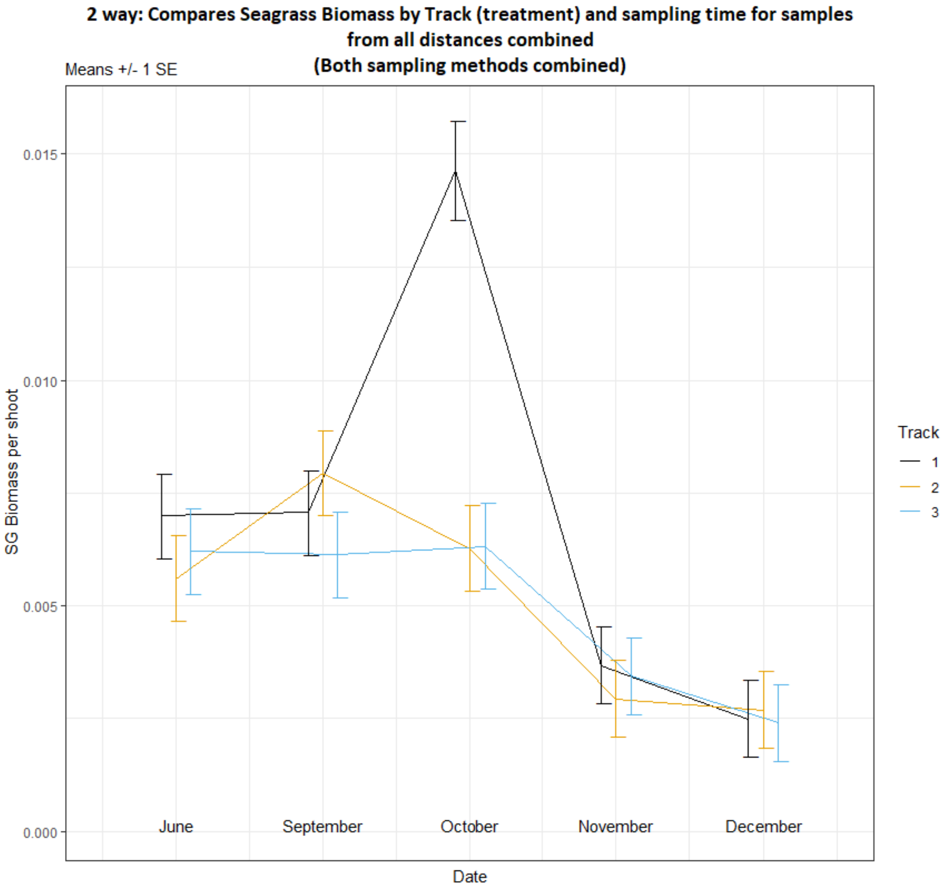
Comparison of seagrass length within track 3 across the distance gradient and time. ANOVA markers show no significant differences between distances from disturbance within Track 3 (Heavy) across all 5 sampling dates which exhibit the seasonal variation.

The two methods of sampling showed similar relationships between leaf length, track, distance and time.

Morphology: Leaf Biomass

Leaf lengths of samples from each measuring station, and collected by both sampling methods, were compared between treatment tracks, distance from disturbance, and sampling times.

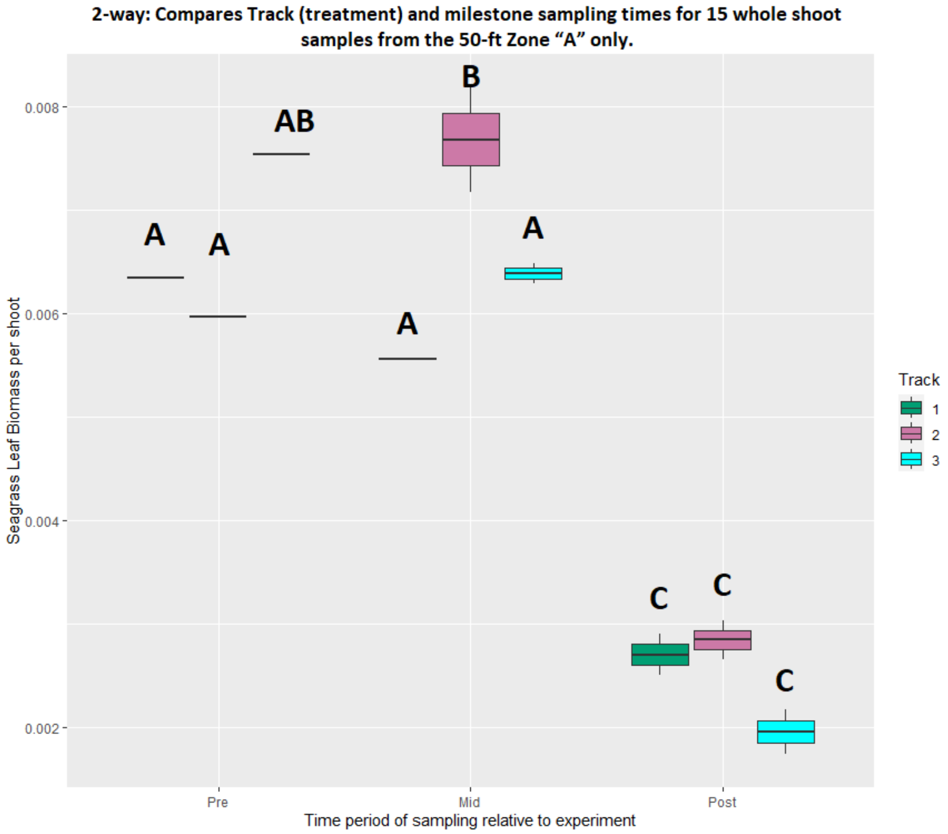
All Data Compared on a Per Shoot Basis



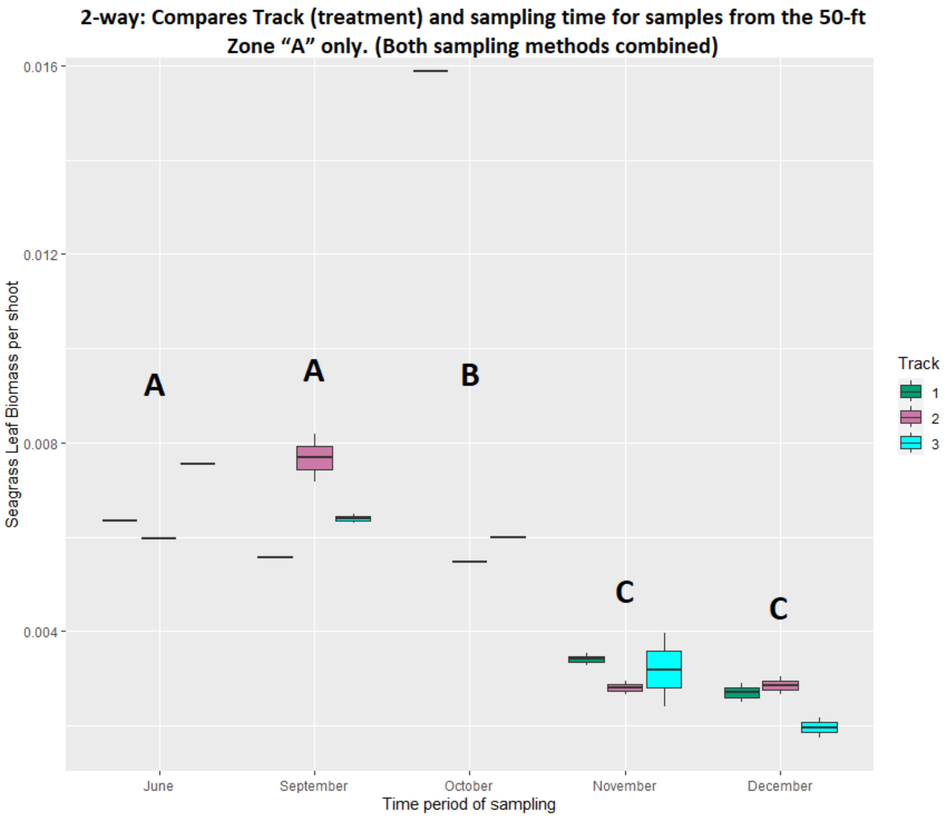
Seagrass biomass did not vary between experimental tracks. The large uptick seen in track 1 could be because of differences in the 2 sampling methods. The seasonal pattern of variation was similar to that observed for leaf length, but there were no significant differences between tracks overall.

Focused Comparisons of Samples from 50-ft Zone "A" for Seagrass Leaf Biomass

The focused comparisons of samples from Zone "A" (50 ft) of the 3 treatment tracks facilitated our objective to compare seagrass leaf biomass between sites with different levels of sediment disturbance.



Focused Zone "A" 50 ft comparison of leaf biomass between the 3 treatment tracks across the milestone sampling times (15 shoot samples) showed no consistently significant differences between tracks. The one exception was a significantly greater biomass for Track 2 (50 ft) during the mid-experiment sampling time. The usual seasonal difference was evident and significant as observed in the other plots.

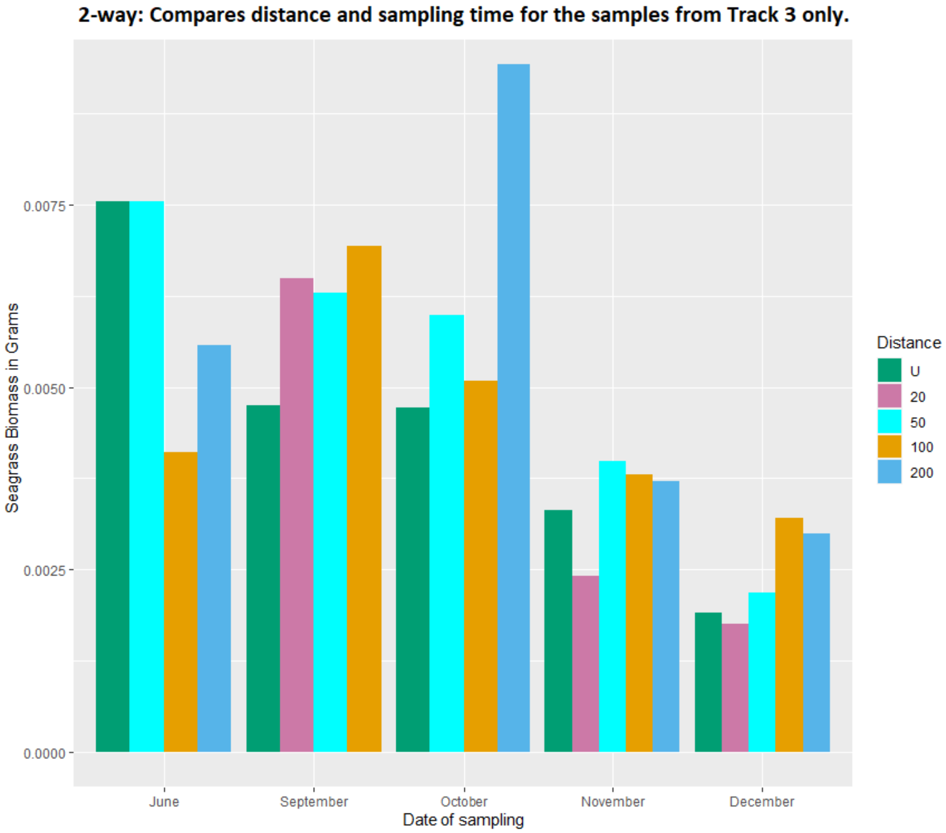


2-way: Compares Track (treatment) and sampling time for samples from the 50-ft Zone “A” only. (Both sampling methods combined)

Focused Zone “A” 50 ft comparison of leaf biomass between the 3 treatment tracks across the all sampling times (both collection methods) showed no significant differences between tracks. The usual seasonal difference was evident and significant as observed in the other plots.

Focused Comparisons of Samples from Track 3 (Heavy Disturbance) With Distance From Disturbance

An alternative focused comparison for samples from the 50 ft heavy disturbance area (Track 3 50 ft) is along the same track across different distances from the disturbance. The distance gradient also corresponds to a depth gradient from ~40 cm to~55 cm.

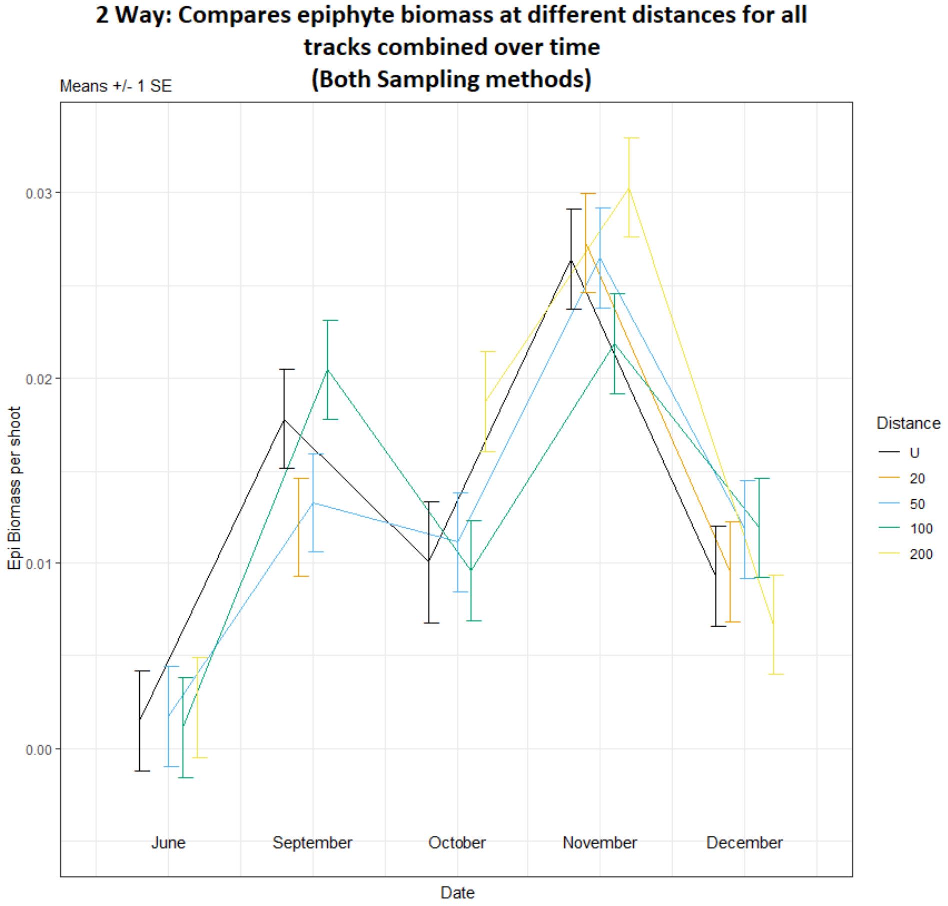


Differences in seagrass biomass within track 3 across the distance gradient were limited and revealed no consistent patterns. This result was for all sampling times combining both sampling methods. The seasonal pattern was observed overall.

Epiphyte Accumulation Metrics

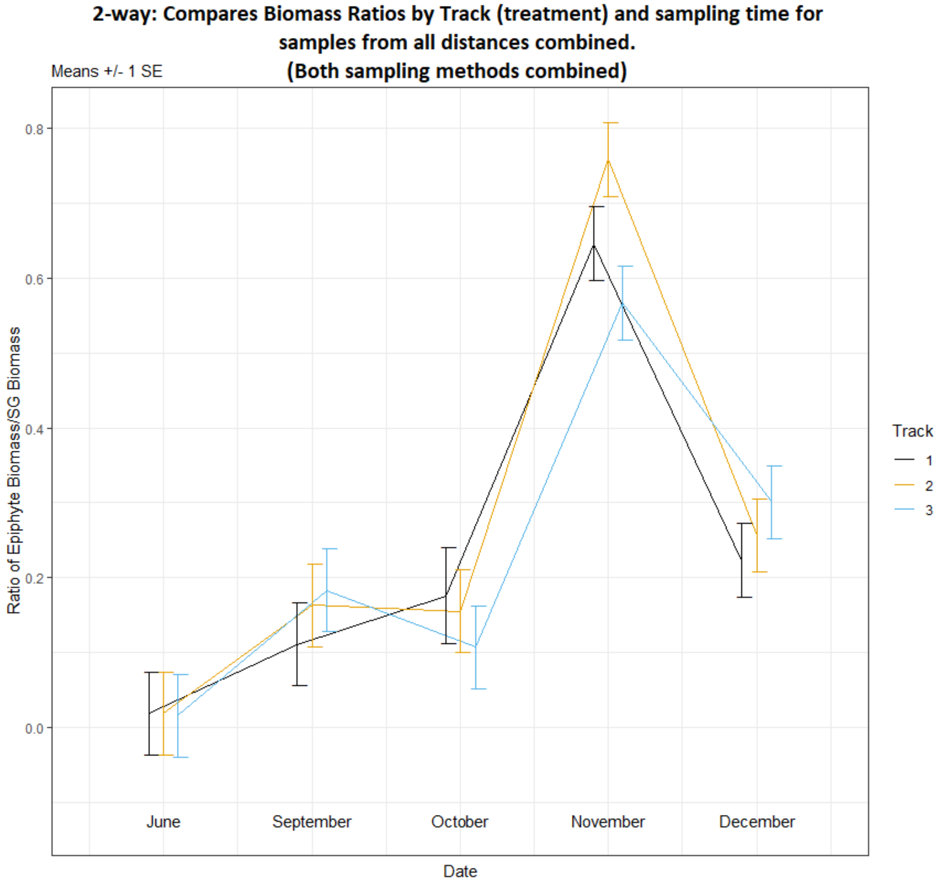
The colonization of seagrass leaves by algae and other diverse organisms is sensitive to environmental conditions, especially light and nutrients. Epiphyte accumulation patterns relative to growth of the seagrass host can serve as indicators, and excessive accumulations can negatively impact seagrass growth and status (Huang et al 2023, Nelson 2017). In addition, the algal composition of the epiphyte community can respond to light and nutrient conditions (Ray et al 2014, Huang et al 2023, Armitage et al 2005). Sediment plumes can potentially impact available light and nutrients, as well as invertebrate grazers of epiphytes, leading to changes in the quantity and community composition of epiphytes, and the way they cover the seagrass leaves affecting the light available to the host leaf. Thus, several epiphyte indicators were measured in this study: epiphyte to seagrass biomass ratio, epiphyte community fluorescence ratio, and percent coverage of the seagrass leaves by epiphytes. These indicators reveal, respectively, the accumulation of epiphytes relative to the seagrass leaf biomass, the relative predominance of green algae vs red algae in the colonizing community, and the relative accumulation on the leaf by areal coverage.

Epiphyte Biomass



Epiphyte biomass did not show any differences or trends for distances from disturbance.

Epiphyte to Seagrass Biomass Ratio

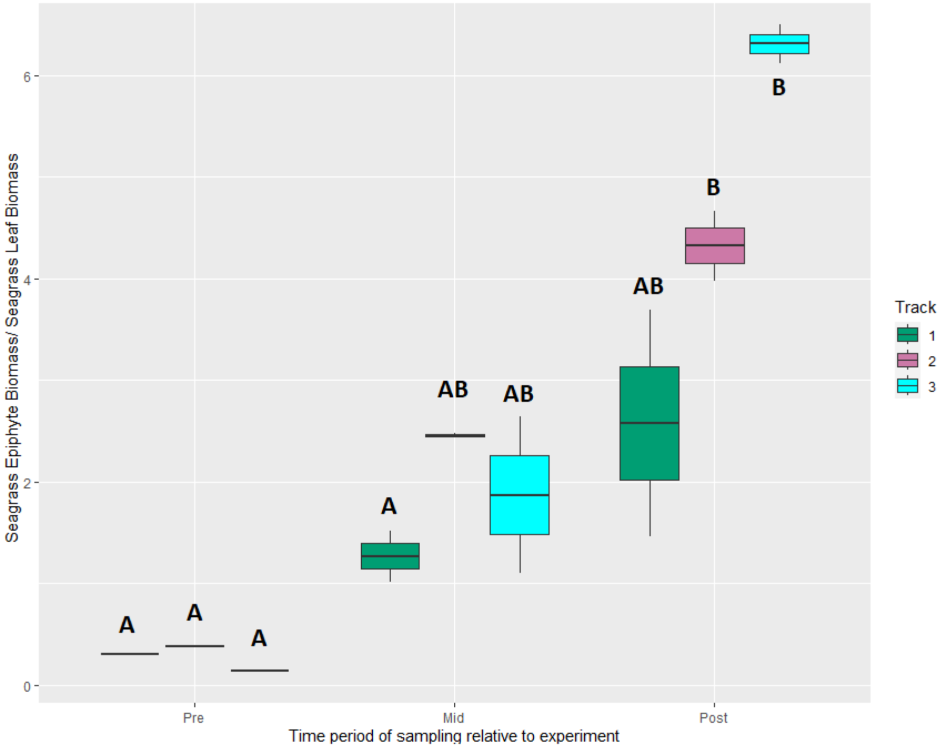


The epiphyte to seagrass biomass ratio is a normalized relative accumulation metric. A clear seasonal pattern is evident where the relative accumulation of epiphytes increases summer through autumn before declining again in December. However, there were no significant differences comparing treatment tracks for all sampling dates and both sampling methods.

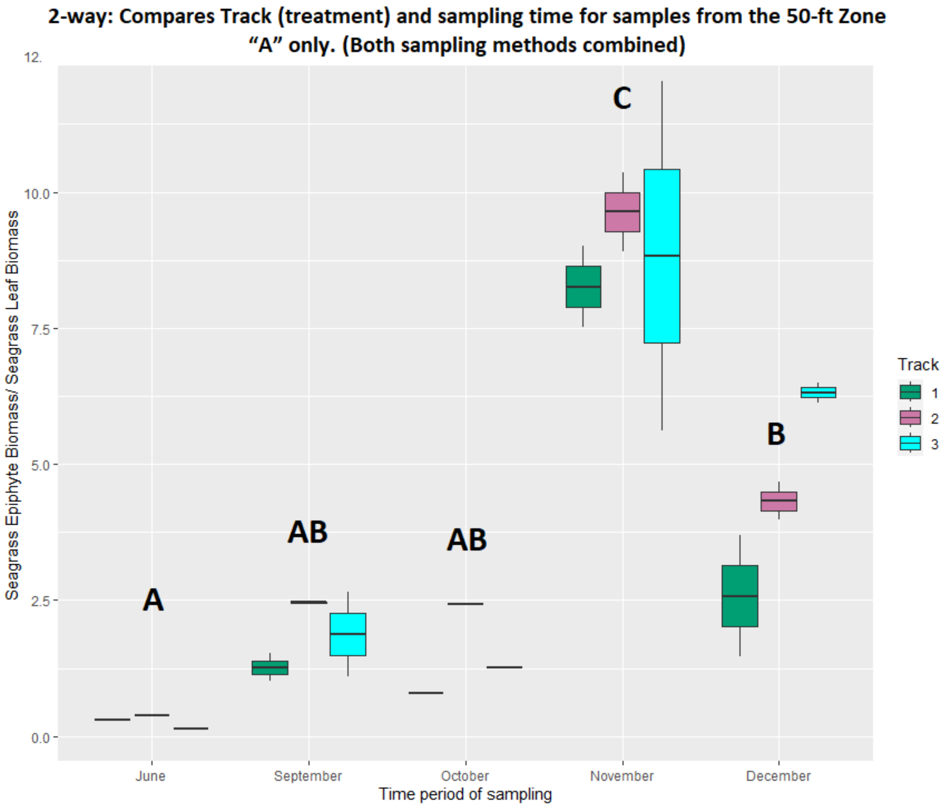
Focused Comparisons of Epiphyte/Seagrass Biomass Ratio for Samples from Track 3 50-ft Zone "A"

A focused comparison of epiphyte/seagrass biomass ratio for samples from different treatment tracks at the 50 ft (zone "A") was made across diferent milestone sampling times using the ring collection method.

2-way: Compares Track (treatment) and milestone sampling times for 15 whole shoot samples from the 50-ft Zone "A" only.



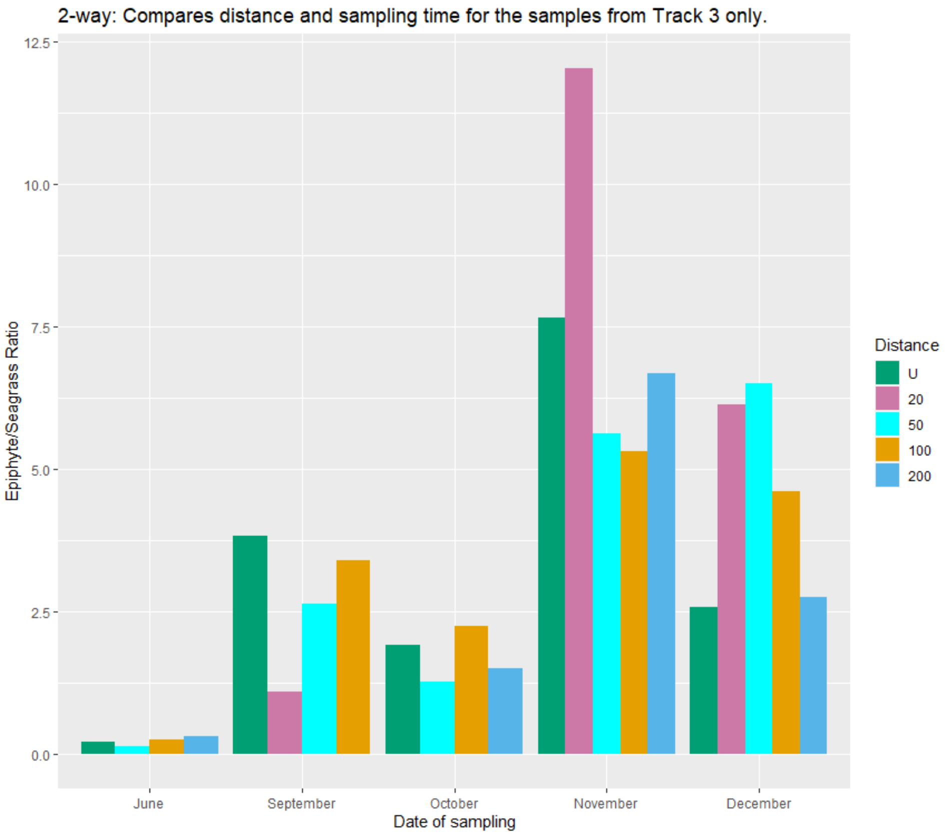
Between-track comparisons of the epiphyte/seagrass biomass ratio within the first 50 ft zone "A" across milestone sampling times revealed no significant differences between tracks at any sampling time points. However, there was a distinct seasonal increase in the ratio, as well as increasing within-sampling period variability with progression through the experiment and seasons. It is possible that cumulative effects or intensity of the disturbances may have played a role in the accumulation of epiphytes on the seagrass blades as the experiment progressed.


2-way: Compares Track (treatment) and sampling time for samples from the 50-ft Zone "A" only. (Both sampling methods combined)

Between-track comparisons of the epiphyte/seagrass biomass ratio within the first 50 ft zone "A" across all sampling times (both collection methods) revealed no significant differences between tracks at any sampling time points. Results are a combination of those in the previous two figures. There was a distinct seasonal increase in the ratio, as well as increasing within-sampling period variability with progression through the experiment and seasons.

Focused Comparisons of Epiphyte/Seagrass Biomass Ratio for Samples from Track 3 (Heavy Disturbance) With Distance From Disturbance

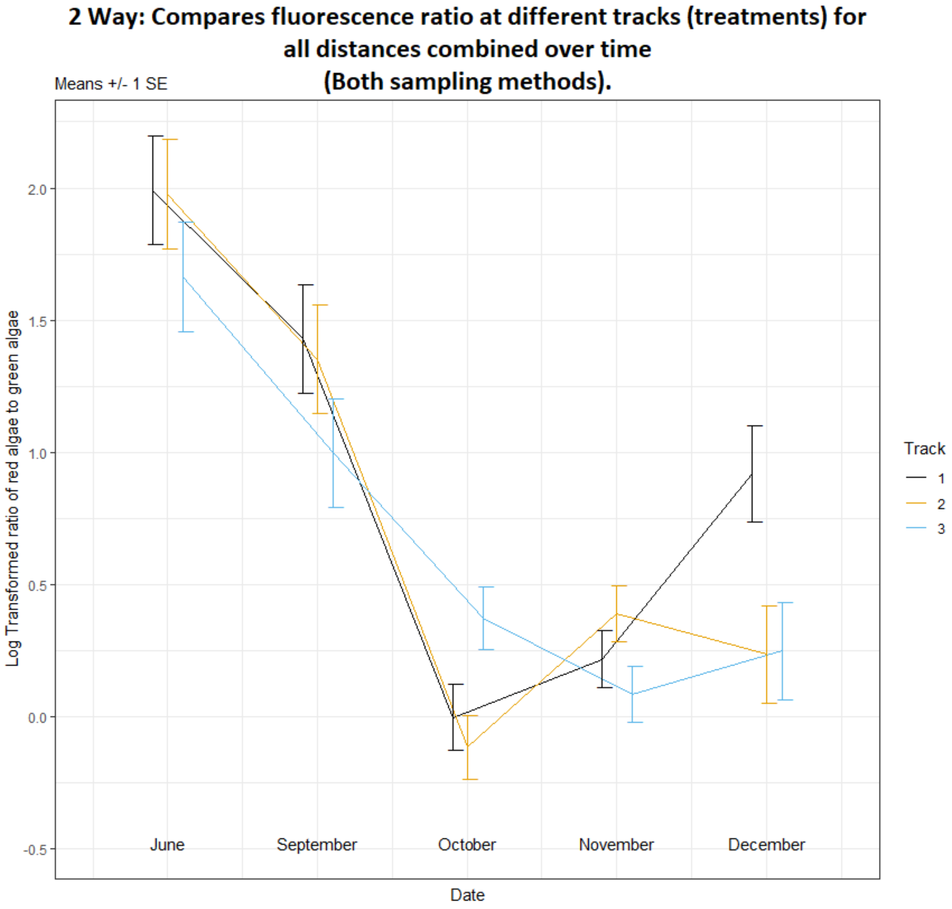
A focused comparison for samples from the 50 ft heavy disturbance area (Track 3 50 ft) was made along that same track for different distances from the disturbance. The distance gradient also corresponds to a depth gradient from ~45cm to ~55 cm.



Comparison of Average Epiphyte/ Seagrass Biomass Ratios (n = 3) within track 3, ordered by distance from disturbance, across all dates of sampling (both methods), revealed no consistent trends or significant differences with distance. The seasonally increasing ratio and level of variability appear again here.

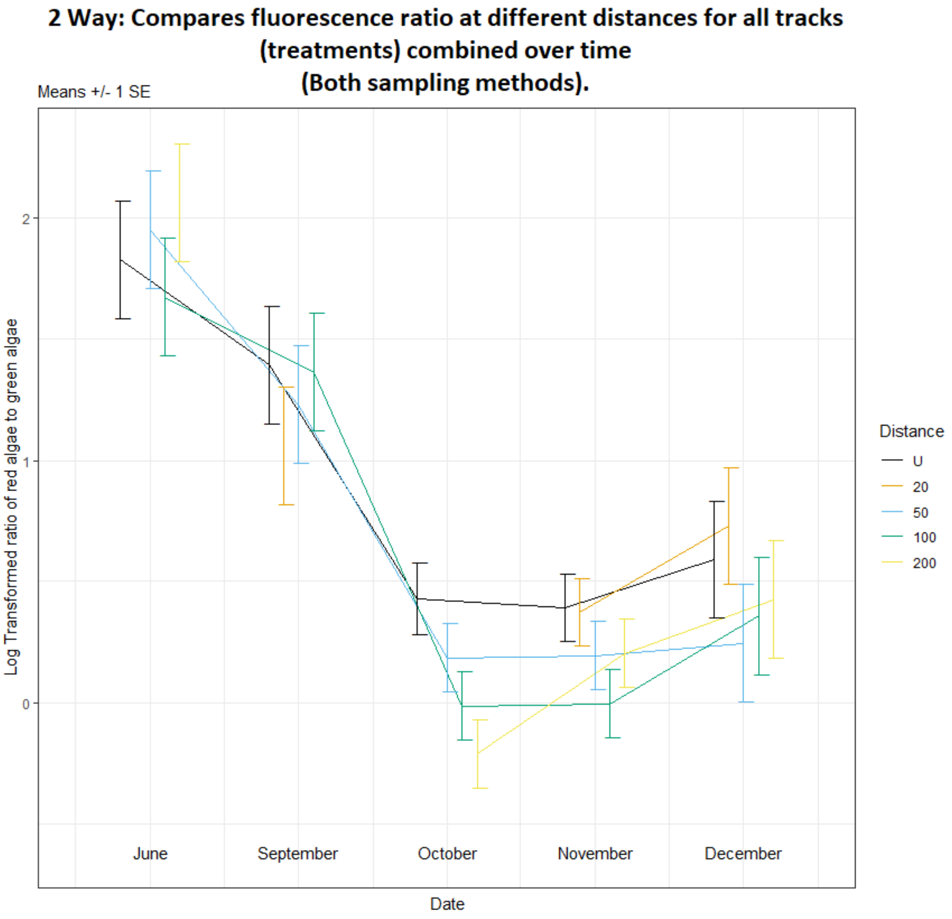
Epiphyte Fluorescence Indicator of Community Composition Changes

The algal community composition, particularly the ratio of red algae to green algae, can change and indicate different environmental conditions. Fluorescence provides a simple indicator of the communities (Ray et al 2014) where the ratio of 576 nm emission/680 nm emission indicates the relative abundances of red and green algae, respectively. Large changes in this ratio suggest a community composition shift. Higher relative ratios indicate an enrichment of red algae relative to green algae overall.



2 Way: Compares fluorescence ratio at different tracks (treatments) for all distances combined over time (Both sampling methods).

Differences in Red/Green epiphyte fluorescence ratio between tracks did not reveal consistent patterns or significant differences between treatment tracks. Note the pattern of decreasing ratio across sampling times (seasons) as the total epiphyte accumulation increases over the same time period. This indicates an increase in the relative proportion of green algal epiphytes as a seasonal pattern.



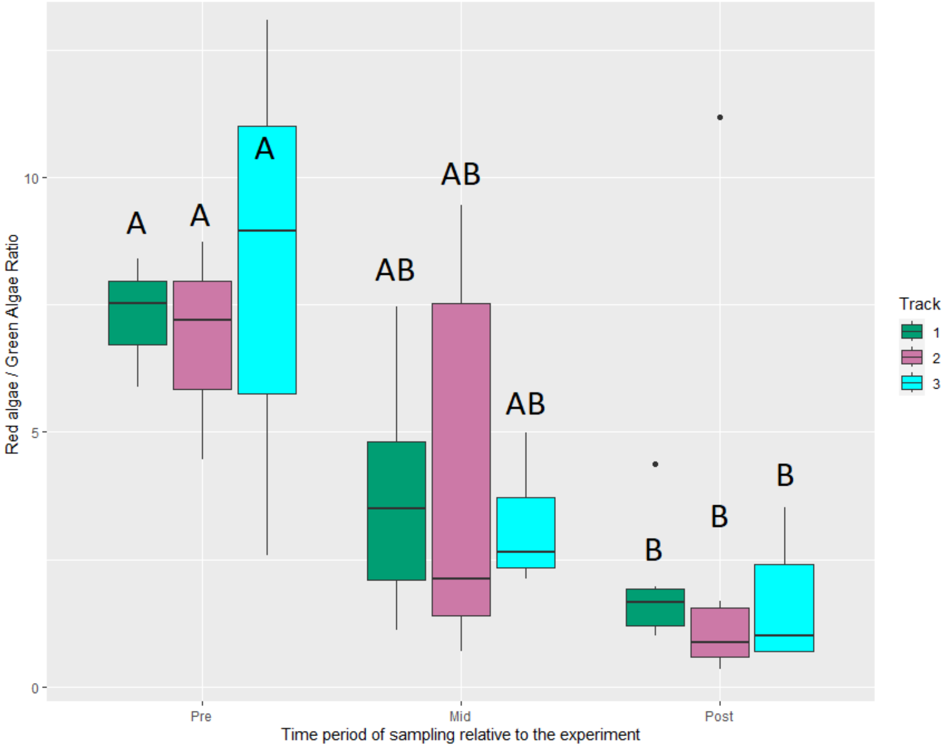
2 Way: Compares fluorescence ratio at different distances for all tracks (treatments) combined over time (Both sampling methods).

Differences in Red/Green epiphyte fluorescence ratio between different distances along all tracks revealed no consistent patterns or significant differences by ANOVA. The seasonally diminishing ratio was consistent at all distances.

Focused Comparisons of Fluorescence Ratio Between Treatment Tracks for Samples from 50-ft Zone “A”

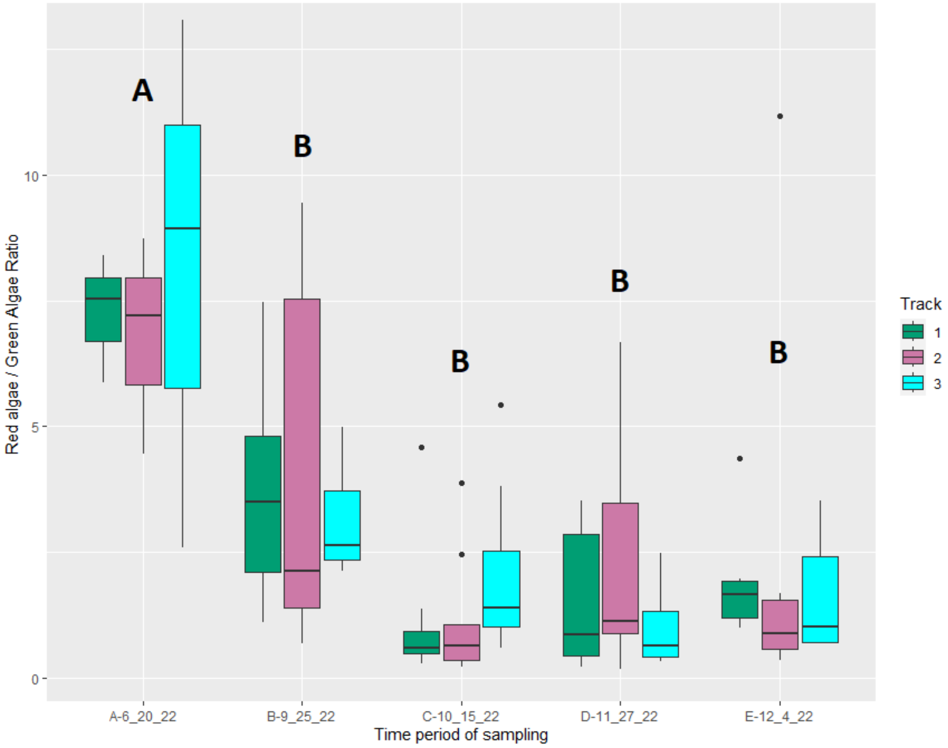
The Red algae/Green algae fluorescence ratio was compared between treatment tracks at the milestone sampling time points (ring collection) in the zone of maximum expected impact (50 ft zone “A”).

2-way: Compares Track (treatment) and milestone sampling times for 15 whole shoot samples from the 50-ft Zone "A" only.



There was no significant difference in the R/G Fluorescence Ratio of epiphytes between tracks for the milestone sampling times (ring collection). The seasonal variation pattern was confirmed.

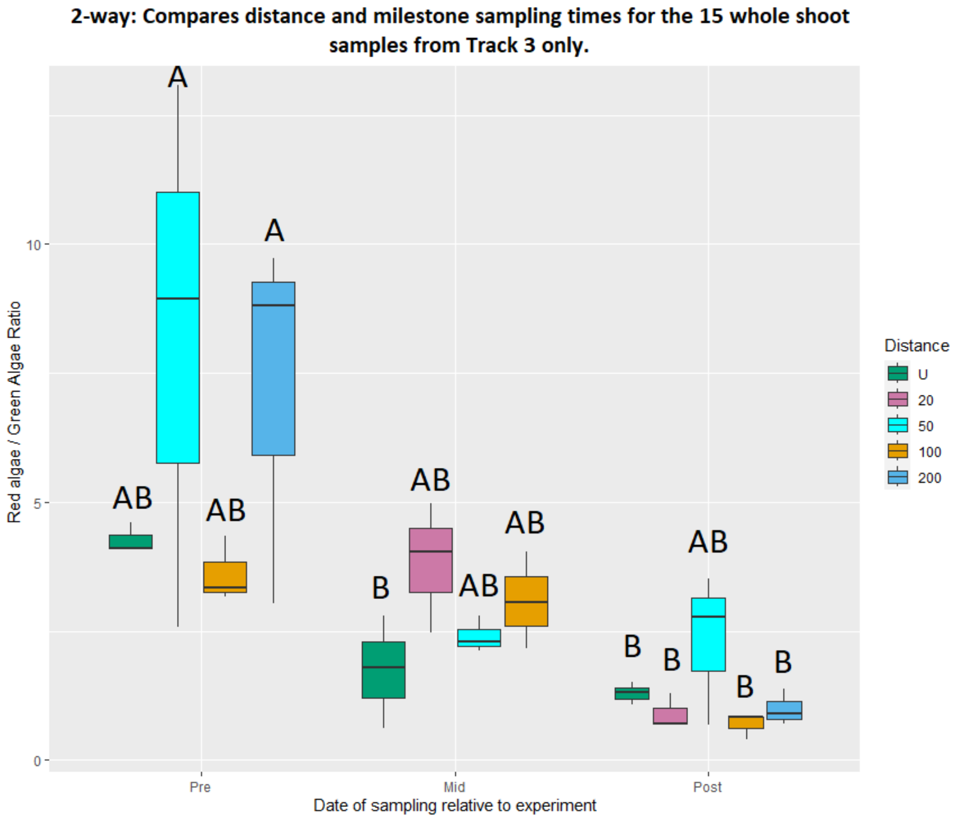
2-way: Compares Track (treatment) and sampling time for samples from the 50-ft Zone "A" only. (Both sampling methods combined)



There was no significant difference in the R/G Fluorescence Ratio of epiphytes between tracks across all sampling times (2 methods). The seasonal variation pattern was confirmed.

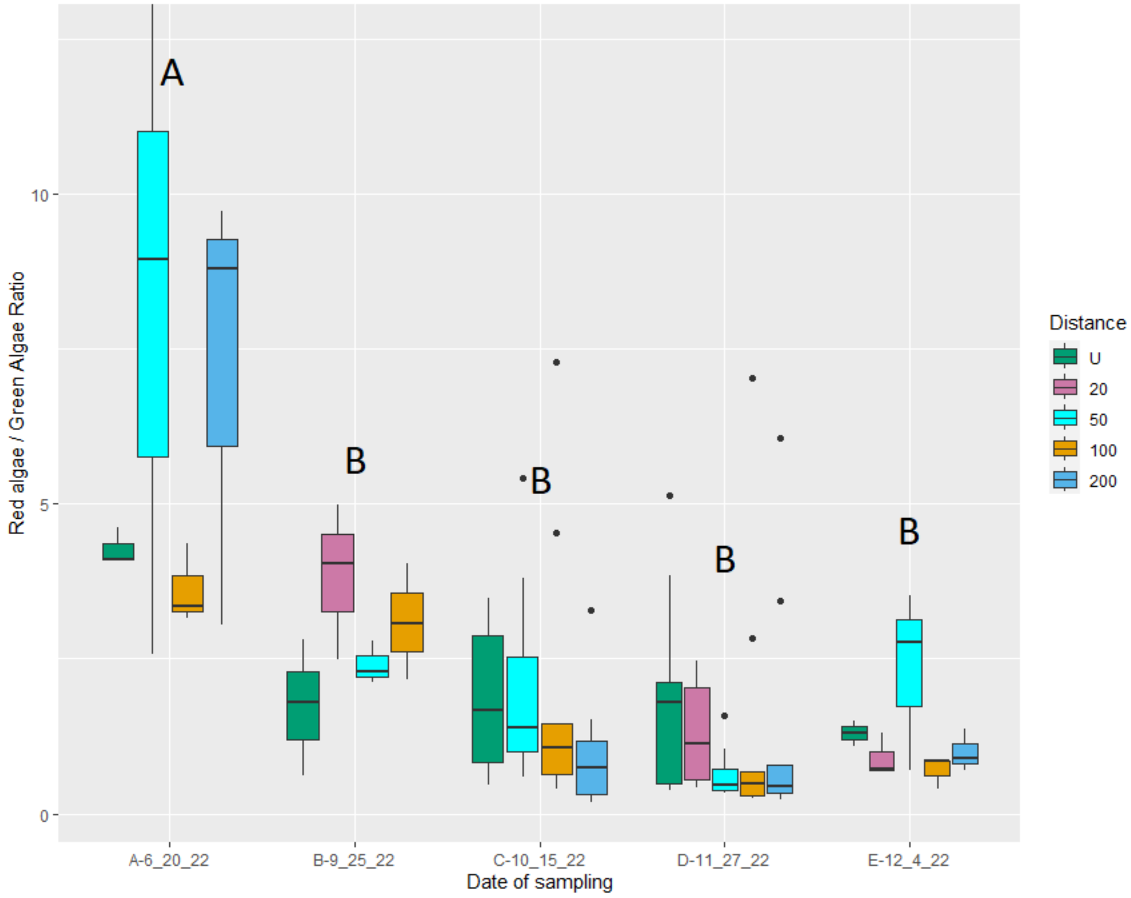
Focused Comparisons of Epiphyte Fluorescence Ratios Along Distance From Disturbance for Samples from Track 3 (Heavy Disturbance) Only

An alternative focused comparison for samples from the 50 ft heavy disturbance area (Track 3 50 ft) is along the same track across different distances. The distance gradient also corresponds to a depth gradient from ~45cm to ~55 cm).


2-way: Compares distance and milestone sampling times for the 15 whole shoot samples from Track 3 only.

Within Track 3 the R/G ratio varied across distances from the disturbance, but there was no consistent trend in the variation and the differences were not significant. Seasonal pattern confirmed for the milestone time points.

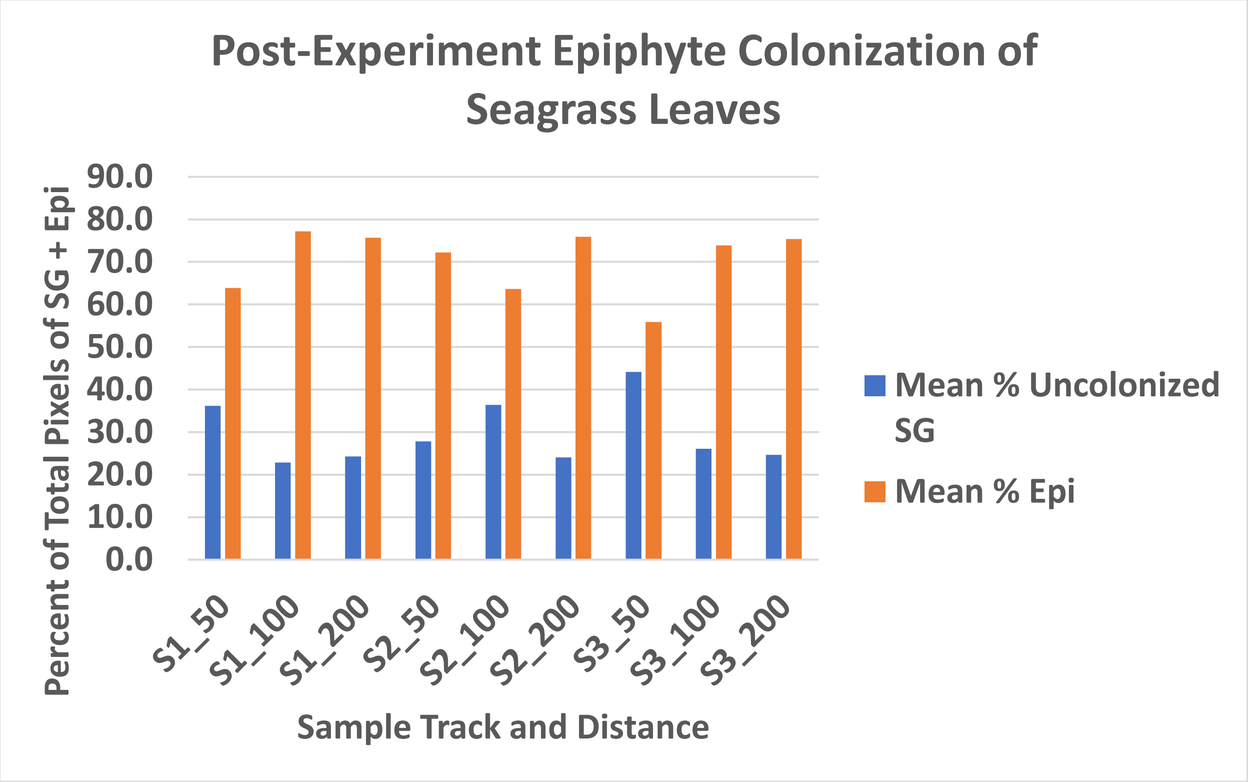
2-way: Compares distance and sampling times for the samples from Track 3 only (Both sampling methods).



Within Track 3 the R/G ratio varied across distances from the disturbance, but there was no consistent trend in the variation and the differences were not significant. Seasonal pattern confirmed for all time points.

Epiphyte Image Analysis

Epiphyte accumulations and their effects on seagrass leaves are caused at least in part by their direct coverage of the leaf and shading of available light. A spatial image analysis of this accumulation and coverage was demonstrated for *Thalassia testudinum* in earlier work (Huang et al 2023). Image analysis was also used here as an alternative measure of epiphyte accumulation. Scanned images of seagrass leaves from the post-experiment sampling were categorized into epiphyte and seagrass classes, which were then quantified by pixel counting.



Plot of comparisons between tracks and distances of relative percent of seagrass leaf colonized by epiphytes, or remaining uncolonized, showed no consistent trends or significant differences in accumulation.

Results Summary Table and Collective Analyses of Data

Each experiment was unique and measurements were conducted in different locations and orientations relative to the sediment disturbances. Multiple experiments were performed on each visit and we had to focus on capturing the sediment plumes which required adapting our experimental design in the field. Ambient conditions varied by day, and often was quite variable during any given day of experiments. Therefore we do not have highly repetitive data for sets of measurements made in the exact same way every time. We provide ranges of observations and metrics for middle values (mean, median) where possible.

SUMMARY TABLE OF SEDIMENT DISTURBANCE MEASURES AT COM AND SEAGRASS SITES

***Settled Solids**: g per 0.0082 m^2 (per day) [Note that Sed Trap value for Lomax North on 1-11-23 was dropped as an artefact]

TSS: Total Suspended Solids (mg/L)

Light Attenuation: % Relative Difference between two loggers 13 cm apart (at seagrass canopy OR at approx. 3 ft depth for deep water measurements); Values discerned by inspection of Relative Difference Plots for individual experiments, comparing attenuation peaks to baseline attenuation.

Diff *Potentially* Assoc w/Activity: An observed change or difference during observation that may or may not be associated with any activities or attributes of the farm

Percent Organic Matter (% OM): Relative % weight loss of dried sediments upon ashing

	N	**Range** (Min to Max)	**Mean** (± Std Dev)	**Median**	**Other Values to Note**
Deep Water COM					
Light Attenuation Overall	4	5% - 80%			Typical Pre-Disturbance Range: 5%-30% Diff *Potentially* Assoc w/Activity: <5% - 10%

Settled Sediments* (g)	16	0.008 – 0.145	0.0523 ± 0.035	0.0471	
(% Organic Matter)	12	(5.47 – 45.68)	(13.33 ± 11.65)	(8.76)	
North	2				**N** 0.026 ± 0.034
(% OM)					(21.84 ± 1.63)
East	3				**E** 0.057 ± 0.061
(% OM)					(20.61 ± 21.72)
South	4				**S** 0.048 ± 0.009
(% OM)					(6.85 ± 1.78)
West	3				**W** 0.066 ± 0.030
(% OM)					(9.02 ± 2.26)
TSS (mg/L)	38	3.653 – 36.471	15.099 ± 8.276	12.362	
(% Organic Matter)	34	(25.806 – 83.333)	(52.019 ± 16.245)	(52.25)	
North	8				**N** 14.616 ± 8.532
(% OM)	7				(52.96 ± 16.93)
East	8				**E** 12.415 ± 5.88
(% OM)	7				(54.73 ± 17.73)
South	8				**S** 14.200 ± 8.63
(% OM)	7				(53.21 ± 15.46)
West	8				**W** 15.12 ± 8.07
(% OM)	7				(56.23 ± 17.65)
Shallow Water COM					
Light Attenuation		5% - 60%			Typical Pre-Disturbance Range: 15%-30% Disturbance Difference: <5% - 20% above pre-disturbance level
Settled Sediments*(g)					
Up/Control	15	0.045 – 3.012	0.880 ± 0.878	0.771	
(% OM)		(0.60 – 9.42)	(3.32 ± 2.27)	(0.281)	
Downstream	86	0.050 – 9.628	1.600 ± 3.094	1.016	
(% OM)		(0.28 – 38.46)	(2.19 ± 4.16)	(1.38)	

TSS (mg/L)					
Up/Control (% OM)	40	8.077 – 216.571 (8.18 – 50.00)	45.008 ± 46.859 (27.99 ± 14.22)	36.346 (24.42)	
Downstream 50 Ft (% OM)	44	10.962 –217.885 (5.05 – 50.00)	55.661 ± 46.987 (24.40 ± 13.48)	39.044 (23.10)	
San Jose Seagrass Site					
Light Attenuation					
Overall Pre-disturbance or Up		5% - 70%			Typical Pre-Disturbance Range: 15%-30%
Down-Current					Disturbance Difference: 5% - 50% above pre-disturbance level
10-1-22 Expt Only Up or Pre-Disturbance	23				10-1-22 Expt Only Up/Pre-Dist: 10% - 30%
Down-Current	22				Down-Current: 25% - 70%
Settled Sediments* (g)					
Up/Control (% OM)	17	0.024 – 3.200 (2.54 – 20.73)	0.695 ± 0.957 (11.03 ± 5.23)	0.210 (10.20)	
Downstream (% OM)	102	0.009 – 9.628 (0.27 – 81.89)	0.753 ± 1.342 (10.67 ± 9.56)	0.217 (8.96)	
10-1-22 Exppt Only					
Up (% OM)	2	0.0283			

Downstream All	11	0.087 – 4.347	0.798 ± 1.232	0.338	
(% OM)		(3.07 – 81.89)	(18.60 ± 24.19)	(9.21)	
TSS (mg/L)					
Up/Control	13	9.143 – 94.510	35.261 ± 30.113	19.157	
(% OM)	10	(19.50 – 50.00)	(30.99 ± 16.86)	(23.45)	
Downstream 50 ft	27	6.538 – 143.250	49.376 ± 44.98	31.321	
(% OM)	26	(5.14 -73.53)	(29.06 ± 20.04)	(24.29)	
10-1-22 Expt Only Up	2	7.692 – 9.811	8.752 ± 1.498		
Down-Current 50 ft	4	12.500 – 42.453	24.014 ± 13.043	20.552	

SUMMARY OF TABLE DATA:

Shallow COM Sites

Based on inspection of the light logger data (Relative Difference Plots) we derived the range and typical values for light attenuation at each site. For the shallow water COM sites, light logger attenuation observations ranged from 5-60% attenuation of light between the top and bottom loggers (top logger situated approximately 3 ft below the water surface). However, the pre-disturbance attenuation typically ranged from 15-30%. Simulated disturbances typically increased light attenuation by an additional 5-20% above the pre-disturbance level. Sediment trap collections varied by the disturbance experiment but ranged from 0.045 – 9.63 g per 0.0082 m^2 per day in the field. Mean values for Control and various downstream collections were 0.88 ± 0.88 g and 1.6 ± 3.09 g respectively. The percent organic matter content of these collections ranged from 0.28-38.46%, with mean values (± Std Dev) of 3.32 ± 2.27 % and 2.19 ± 4.16 %. In addition, despite a high degree of variability, fewer sediments were typically collected at greater distances, and the % organic matter generally increased with distance from the disturbance.

Similar phenomena were observed for TSS samples at the shallow sites. TSS values ranged from 8.08-217.89 mg/L, with mean values of 45.01 ± 36.35 mg/L and 55.66 ± 46.99 mg/L observed for upstream controls and downstream collections at 50 ft, respectively. Observed % organic content of TSS ranged from 5.05-50% with mean values (± Std Dev) of 27.99 ± 14.22 % and 24.4 ± 13.48 % observed for Control and downstream samples, respectively. TSS values generally decreased with distance from the disturbance but these values were highly variable with conditions and experimental setup. If dense inorganic sands settle out quickly (near the disturbance source) it would be expected that remaining less dense organic matter might travel farther before settling, and that the character of the sediment plume would change by having an increased % organic content. Accordingly, light attenuation diminished with distance from the disturbance. However, there was too much variability in TSS and sediment trap data to either support or refute this prediction.

The nature of the measured characteristics of the sediment plumes associated with disturbance activity was highly dependent on hydrology (wind and current), ambient turbidity (generally high in these shallow, exposed and unvegetated sites), and position of the measurement instruments. It was difficult to predict all factors and place instruments optimally. Clearly, sediments will be directed in different directions from their source depending on these changeable conditions. But these measurements capture what we believe to be realistic short term suspended sediment characteristics. The frequency, duration, location and potential long term impacts remain unknown.

Deep COM Site

The deep water commercial COM operation observed in Copano Bay (TX Oyster Ranch) required adaptations to our observation and measurement procedures. Measurements were made from floating cages positioned on 4 sides of the farm. In the absence of bottom disturbances at these deeper sites, measurements captured a combination of ambient suspended and settling sediments plus any biota colonizing the cages which might be knocked off during handling (along with any water-column sediments that became entrained in such biofilms) plus any biodeposits that might reach the measurement stations.

Inspection of the light logger data (Relative Difference Plots) for the deep water COM site showed light logger attenuation values that ranged from 5-80% attenuation of light, with the highest values obtained from deeper in the water column. The pre-disturbance (in this case morning values before farm work started, but also when water conditions were generally calmer) difference, between the top and bottom loggers (top logger situated approximately 3 ft below the water surface) typically ranged from 5-30% attenuation. Afternoon conditions, following a combination of farm work and generally increasing winds and currents, were most often 5-10% greater than the morning value.

Deep water COM sediment trap collections ranged from 0.008 – 0.145 g per 0.0082 m^2 per day in the field. The mean value for all collections was 0.052 ± 0.035 g and the median value was 0.047 g. The percent organic matter content of these collections ranged from 5.46-45.68%, with a mean value (± Std Dev) of 13.33 ± 11.65 %. The Mean deep COM overall sediment trap collection value of 0.052 ± 0.35 g was 30-fold lower than the mean sediment trap collection values observed at the shallow COM downstream sites, and 16.9-fold less than observed at the shallow up-current control sites. There was an interesting distinction between collection sites at the deep COM when comparing % organic matter of the collected sediments. The North site was at least twice as high as for the South and West sites. The value for the East site was quite variable, with a mean value similar to North, but a median value more like South and West. The means and medians were very similar for all other sites.

Deep water TSS values ranged from 3.65-36.47 mg/L, with a mean value of 15.1 ± 8.28 mg/L. Observed % organic content of TSS ranged from 25.81-83.33% with a mean value (± Std Dev) of 52.02.99 ± 16.25 %. These TSS values are generally lower than observed for the shallow COM sites, but the % organic matter was greater at the deep water site. Unlike the sediment trap values, the mean TSS values for each measuring location were very similar, the North site did not stand out in this case.

It was not possible for us to make any measurement attribution directly to the farm work as the observed work was often scattered around the large farm area and occurred rather quickly over short periods of time in the mornings and afternoons. Thus any detectable light and sediment trap measurement changes could only *potentially* be due to some combination of farm work and ambient water quality changes with changing winds. So these numbers, *at best*, could only provide an upper detectable limit. There were instances where our measurement platform was within 50 ft of cages that were worked-on, yet we could not discern any measurement changes. Material released from the cages after, eg. bag shuffling, was captured by aerial UAS imagery as a sediment plume drifting a short distance and then dissipating within minutes, but it was not discernable in our measurements. It would take a much larger number of measurement stations in a gridded array around the farm to improve the probability of capturing these events in measurements. The UAS imagery and subsequent image analysis may be better suited to capturing the extent of plume generation.

San Jose Seagrass Site

Simulated sediment plumes of varying intensities were applied to the seagrass study site over the ~ 5-month experimental period. Totals of 811 p-m and 455 p-m were accumulated in zones “A” (0-50 ft) at Tracks 3 (heavy) and 2 (light), respectively.

Relative Difference Plots of light logger data showed that light attenuation at San Jose experiments ranged from 5% - 70%. Upstream controls or pre-disturbance baseline attenuation was most typically 15%- 30%. Simulated disturbances increased light attenuation by an additional 5% - 50% above the pre-disturbance level. In the best studied of our experiments (10-1-22), Upstream and pre-disturbance baseline attenuation varied from 10% - 30%, and down-current logger poles recorded attenuations ranging from 25% - 70%, translating into sediment plume-attributable attenuations of 15% to 40%.

Sediment trap collections overall at San Jose ranged from 0.01 – 9.63 g per 0.0082 m^2 per day in the field. Mean values for Control and various downstream collections were 0.70 ± 0.96 g and

0.75 ± 1.34 g respectively. The percent organic matter content of these collections ranged from 0.27-81.89%, with mean values (± Std Dev) of 11.03 ± 5.23% and 10.67 ± 9.56%. For the 10-1-22 experiment by itself, the Control sediment collection was 0.028 while the mean down-current collection was 0.798 ± 1.232 with a mean organic content of 18.6 ± 24.2%. Sediment collections diminished with distance from the disturbance, but the majority of near-disturbance suspended sediments were settled out in the first 50 ft (see figure below for 10-1-22 experiment).

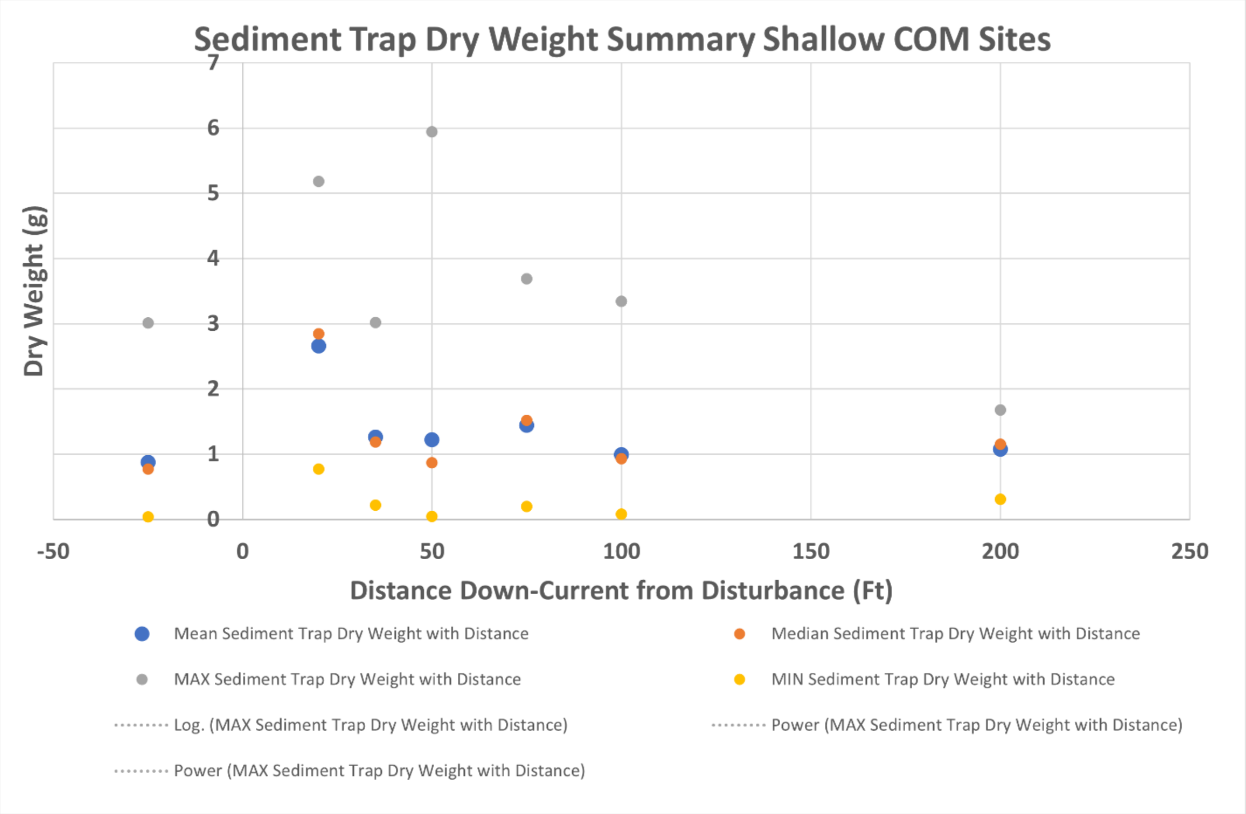
TSS levels overall at San Jose ranged from 9.14 – 143.25 mg/L, with mean values of 35.26 ± 30.11 mg/L and 49.38 ± 44.98 mg/L observed for upstream controls and downstream collections at 50 ft, respectively. Observed % organic content of TSS ranged from 5.14-73.53% with mean values (± Std Dev) of 30.99 ± 16.86 % and 29.06 ± 20.04 % observed for Control and downstream samples, respectively. For the 10-1-22 experiment by itself, mean TSS values were 8.75 ± 1.5 mg/L and 24.01 ± 13.04 mg/L at the Control and downstream sites. Overall TSS values generally decreased with distance from the disturbance but these values were highly variable with conditions and experimental setup. If dense inorganic sands settle out quickly (near the disturbance source) it would be expected that remaining less dense organic matter might travel farther before settling, and that the character of the sediment plume would change by having an increased % organic content. Accordingly, light attenuation diminished with distance from the disturbance. However, there was too much variability in TSS and sediment trap data to either support or refute this prediction.

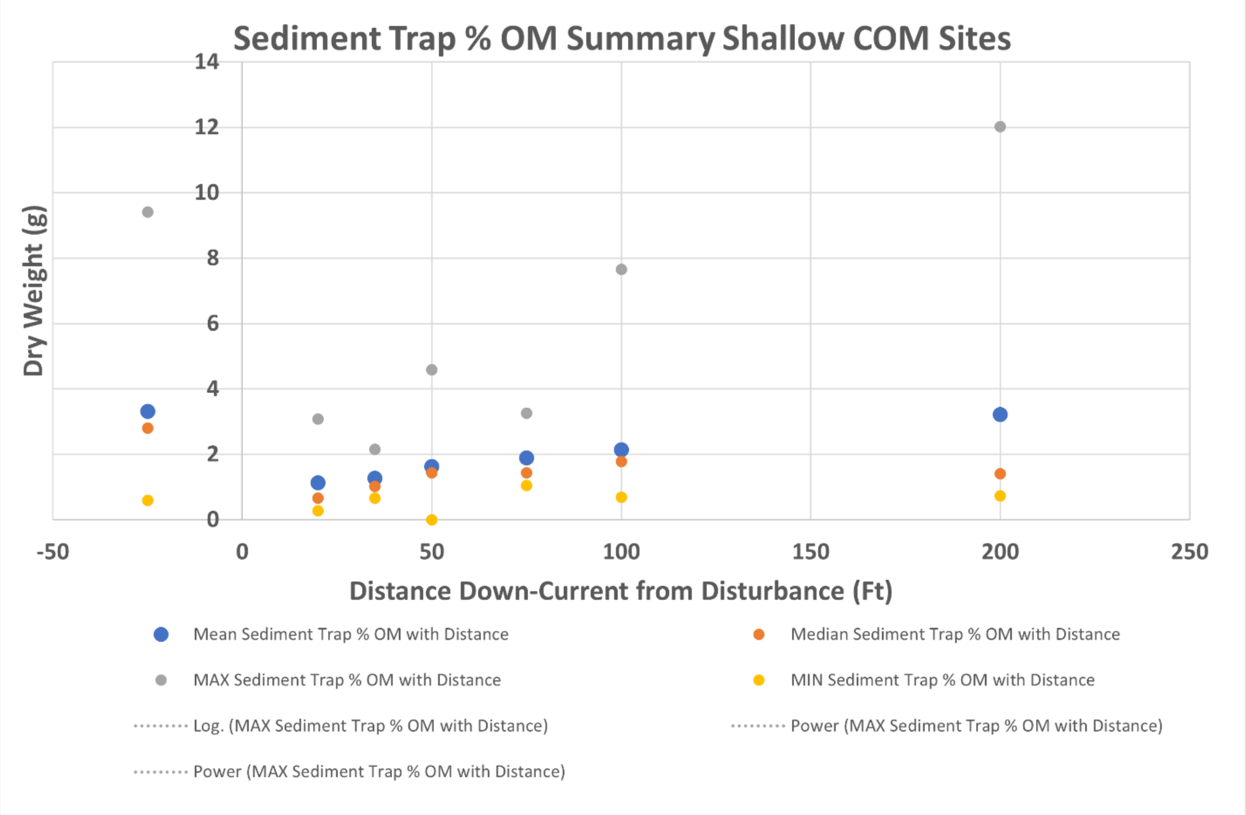
ANALYSES OF DATA COLLECTIVELY:

Shallow COM Sediment Plume Metrics

The sediment trap dry weight collection summary data, for experiments at all 3 shallow COM sites combined, were plotted relative to the distance gradient from the disturbances. The large blue dots represent the mean values from all experiments, the orange dots represent median values, and the gray and yellow dots, respectively, represent the maximum and minimum values (range). Points plotted at -25 ft are the upstream or control values. All of these summary values trace a similar trajectory. Mean and median values are nearly identical, and are approximately 3-fold higher at 20 ft down-current compared to upstream/control values. At 35 – 50 ft distance the values are only slightly greater than control values, and they are the same at 100 ft. and 200 ft.

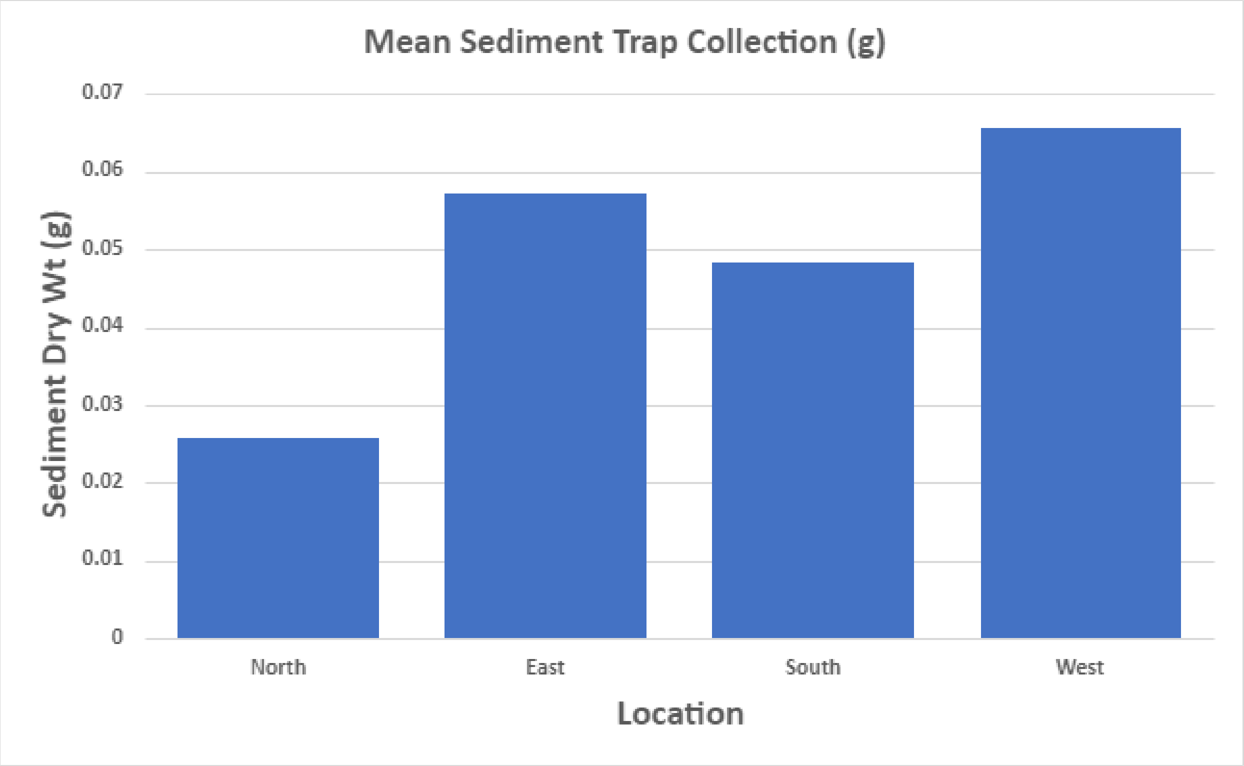
The next plot represents the % Organic Matter (% OM) summary statistics for these samples with the same color scheme. Values at 20 ft down-current are less than half compared to the controls, and the increase over distance until they are nearly identical to control values by 200 ft distance. Similar trajectories are observed for the median and maximum values as well. Thus the collective mean sediment trap values for the shallow COM experiments exhibit the aforementioned expected behaviors that would be consistent with more sandy, more inorganic suspended sediments settle rapidly within 35 – 100 ft while less dense particles with greater organic content travel farther before largely settling by 200 ft.

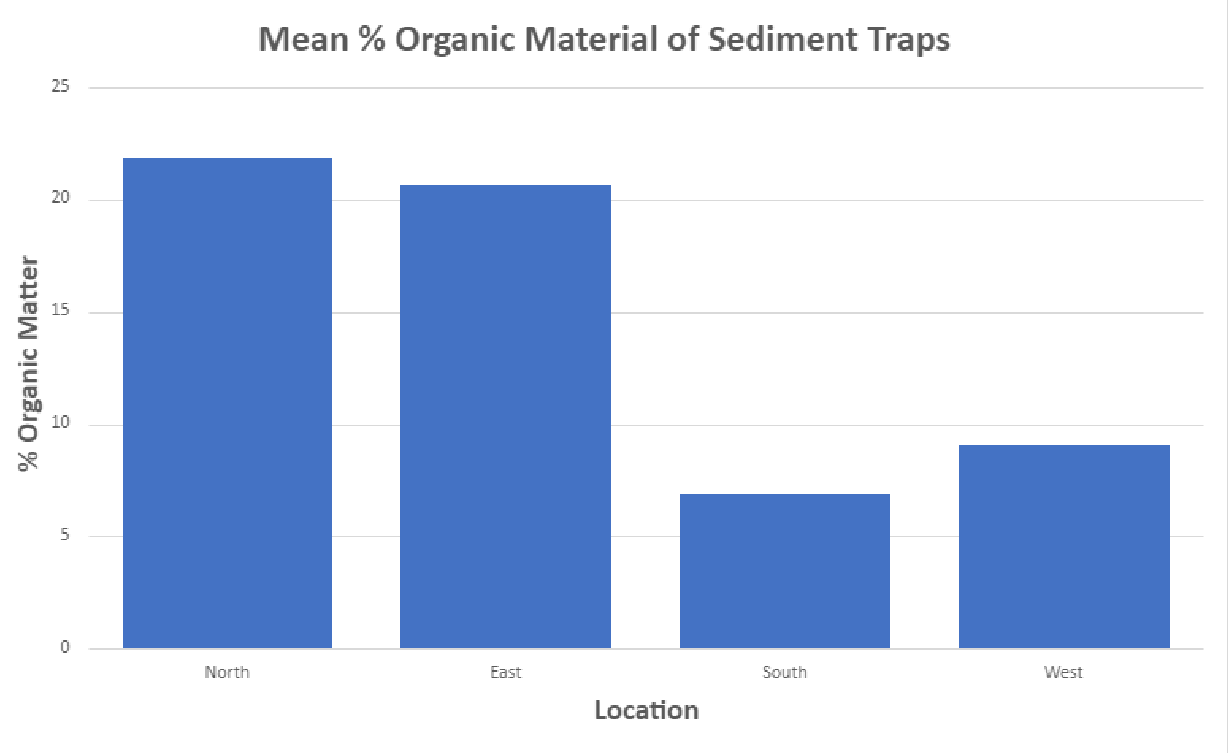




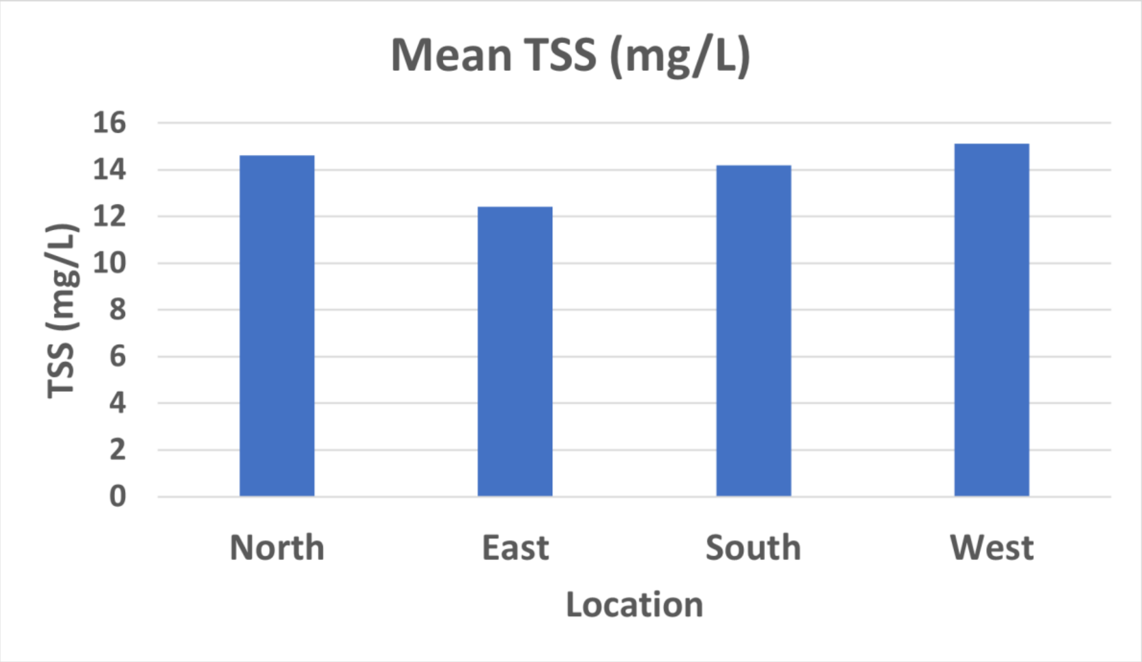
Deep COM Sediment Plume Metrics

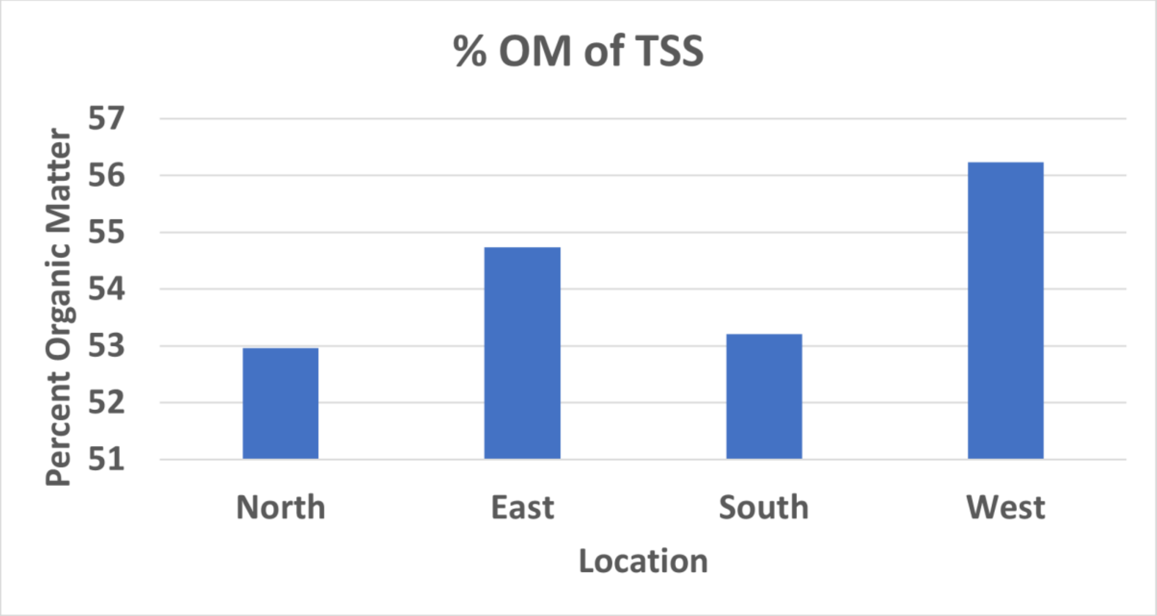
Plots of the sediment trap dry weight collection and % OM summary data for each location at the deep water COM depict an unclear understanding of sediment plumes at the active deep water farm. The sediment trap dry weight collection values at North were rather similar to those at the other locations except for the 1-11-23 experiment which had a value almost 2 orders of magnitude higher than for the other sites that day. This is presumably an artifact of the trap being near the bottom, and if that outlier is removed, the North overall mean sediment collection value (0.03 g) is about half that at the other sites (0.05 – 0.07 g) and probably more representative of that site. In that case, analyzed collectively, the data show the lowest collections at the North side. But these samples also showed the highest % OM in comparison to the other sites. The East site in particular had highly variable data with a wide disparity in the mean and median values for sediment trap collections, and a high % OM similar to the North site (>20% compared to <10% at the South and West locations). The mean overall values contrast those for individual experiments which often showed location-specific differences at this farm. We attribute this to variable weather and current patterns.


Mean Sediment Trap Collection (g)
Sediment Dry Wt (g)
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0
North
East
South
West
Location


Mean % Organic Material of Sediment Traps
% Organic Matter
25
20
15
10
5
0
North
East
South
West
Location

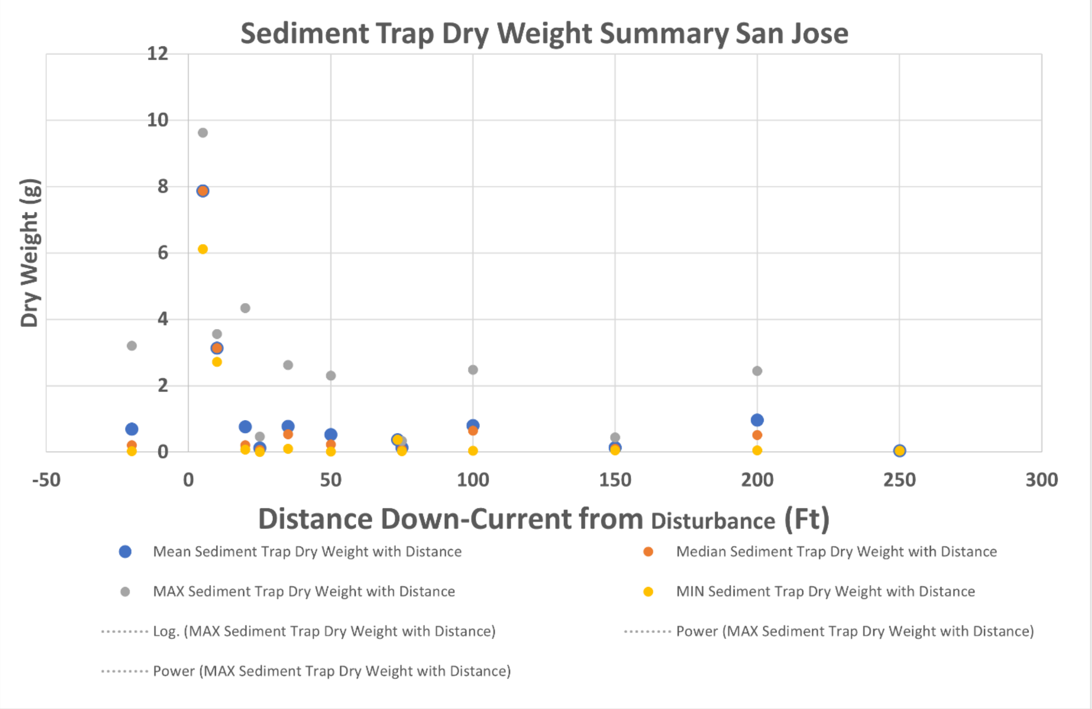
The TSS assay at the deep COM site tells a much different story. TSS levels at the deep COM were much lower than those observed at the shallow COM sites (2.3 to 5.6-fold lower), but there were no significant differences overall between the locations within the deep COM farm. The TSS % OM was likewise not significantly different between locations, but the overall mean TSS % OM value was approximately 2-fold greater than at the shallow COMs (~52% vs ~26% respectively). Although some of the differences observed for experiments at the shallow COM sites vs the deep COM site could be attributed to the simulated disturbances applied at the shallow sites, the increased sediments and decreased % OM for both TSS and sediment trap assays were also observed for the undisturbed or upstream control locations.



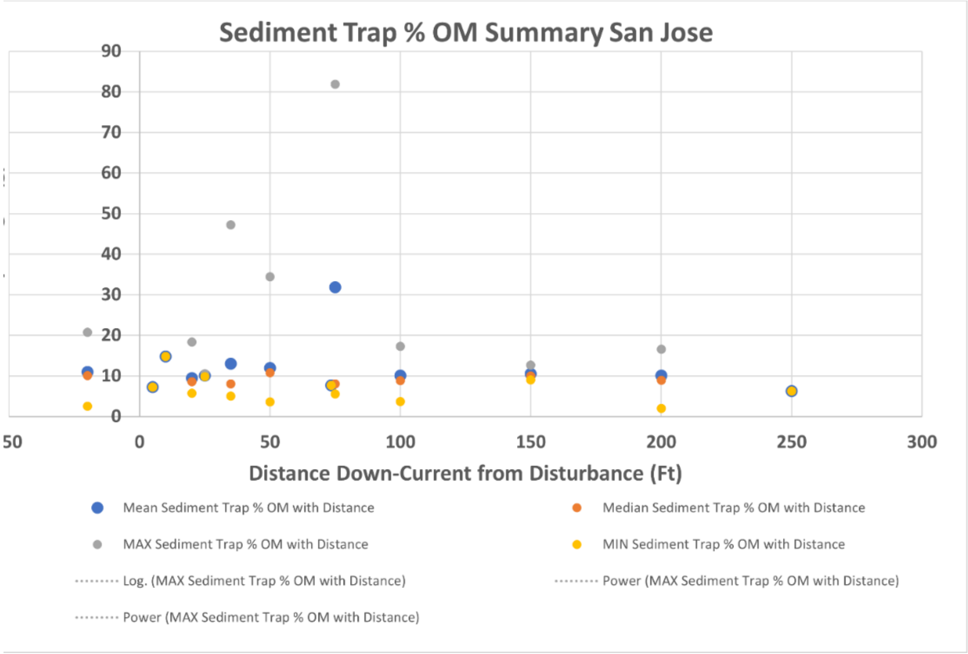


San Jose Seagrass Site Sediment Plume Metrics

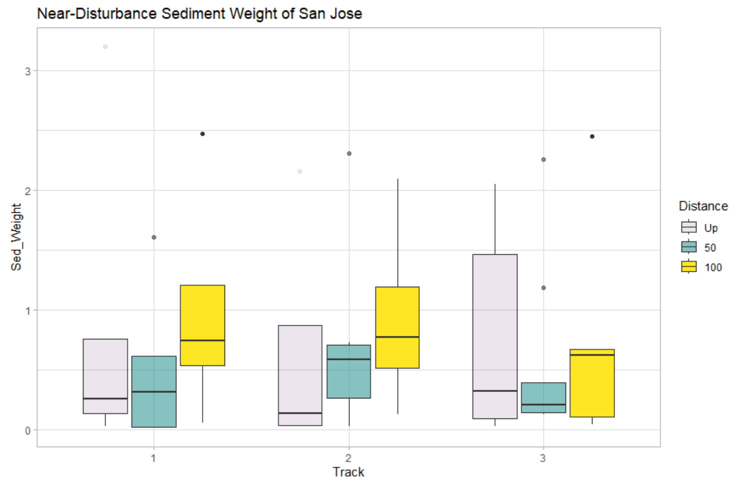
The sediment trap dry weight collection summary data, for all seagrass site experiments combined, were plotted relative to the distance gradient from the disturbances. The large blue dots represent the mean values from all experiments, the orange dots represent median values, and the gray and yellow dots, respectively, represent the maximum and minimum values (range). Points plotted at -20 ft are the upstream or control values. All of these summary values trace a similar trajectory. Mean and median values are close and are approximately 8 fold higher at 5 ft down-current, and 3-fold higher at 10 ft down-current compared to upstream/control values. At 15-35 ft distance the values are similar to the control. Suspended sediments did not travel very far before settling when measured by weight. This overall average does not reflect all conditions as the 10-1-22 experiment clearly demonstrated sediment plumes reaching 300 ft and settleable solids moving up to 100 ft.



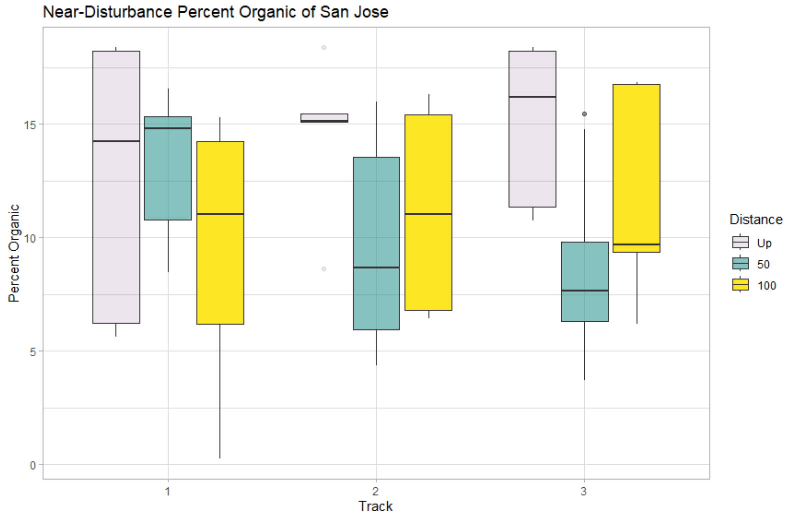
The next plot represents the % Organic Matter (% OM) summary statistics for San Jose sediment plumes overall, using the same color scheme. There were no discernable trends in the this overall % OM data.



It is important to revisit the treatment track comparisons with the collective San Jose data. The relatively short travel distance of measurable suspended solids here may have confounded our experimental design to make comparisons between treatment tracks at 50 ft and greater distances. Data obtained from very short distance measurements (< 35 ft) were underrepresented in our dataset, so the tracks must be compared using the originally established sampling stations. A focused comparison for the presumed most-impacted zone "A" 50 ft data (near-disturbance plots below) does not reveal any significant differences in sediment trap collections between treatment tracks for the Up, 50, and 100 ft measuring stations within tracks. But as can be seen from the plots above, most measurable sediments from the generated plumes would have settled before traveling to the 50 ft distance. The 10-1-22 experiment, which had a strong current driven by aligned tidal and wind forces, provides a notable exception to this overall summary.

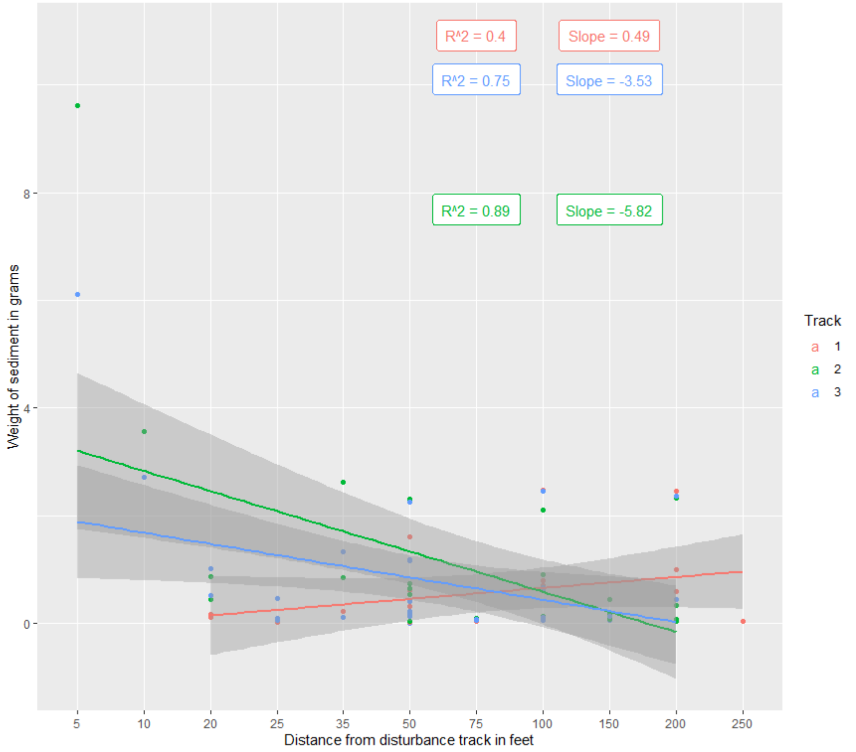


Likewise, there were no significant differences comparing % OM between the 3 tracks at the near-disturbance distances (below). However, at Track 3, the % OM was significantly lower at 50 ft vs Upstream (p=0.01), and for Track 2 the difference was marginally significant (p = 0.094), which is what we would expect for a sediment plume with a relatively greater composition of sands.

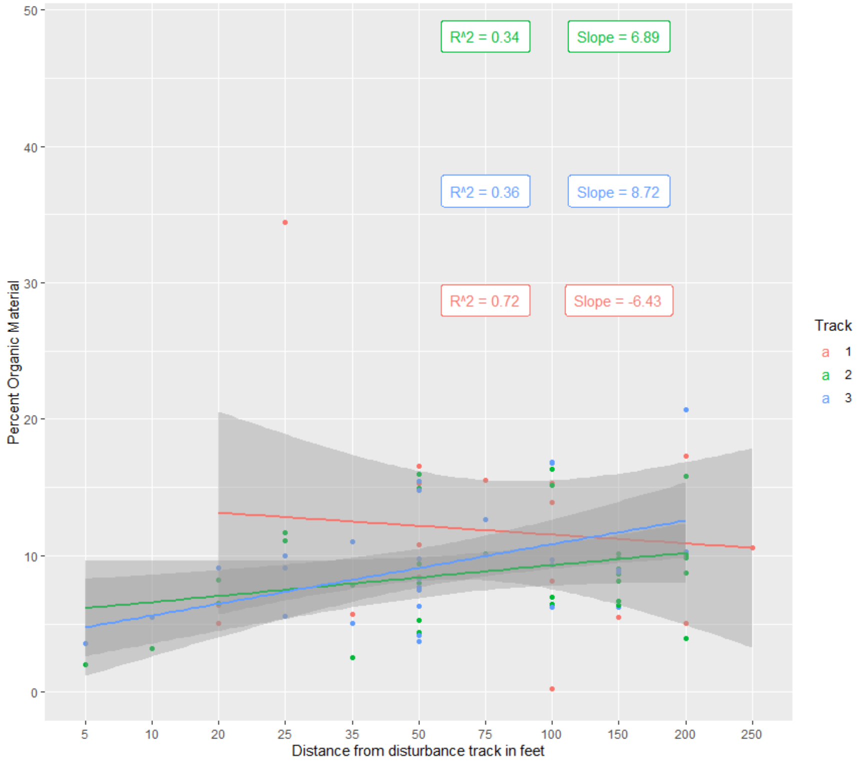


Exploring the collective between-track comparisons further, the plots below show sediment trap collections and % OM for distance from the disturbance, along with 95% confidence intervals. For both sediment dry weight and % OM, there were different trends (slopes) observed for the control Track 1 and the disturbance Tracks 2 and 3. Despite the moderate correlations and different slopes, these plots were not significantly different.

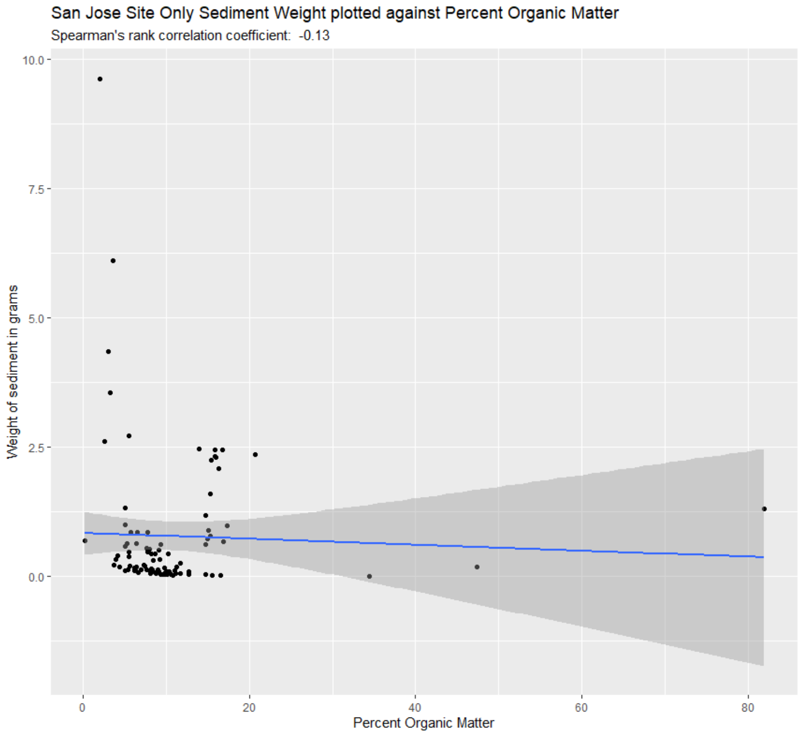
San Jose Site Only Sediment Weight plotted along distance gradient tracks outlined by color



San Jose Site Only Percent Organic plotted along distance gradient tracks outlined by color (Sediment



A number of observations left the impression that greater sediment trap weight collections had lower % organic content. This was tested in a correlation plot of sediment trap dry weight vs % OM below. The correlation was very weak (-0.13) and negative for the San Jose data overall.



Additional Data Comparisons and Correlations

Work to understand the project data is anticipated to continue over the next year as student researchers complete their projects to further analyze the dataset collectively as a whole and in correlation with light attenuation data. A variety of additional approaches will be used, such as Primer+Permanova for analysis of covariance, and tools available on Cyverse-Bisque for image analyses.

Dissemination, Outreach and Stakeholder Input

Dissemination

Project findings will be widely disseminated to the scientific community through a variety of means, including peer-reviewed publication, scientific or general public presentations, conferences, websites and public data repositories. In all cases, the financial support of TGLO will be acknowledged. The draft report has been disseminated to colleagues at TPWD, MANERR, COM operators and COM researchers in a request for comments and stakeholder input. This finalized report has been sent as a followup, and it will be shared with the TX Seagrass Monitoring Working Group at the next available meeting.

Data Availability

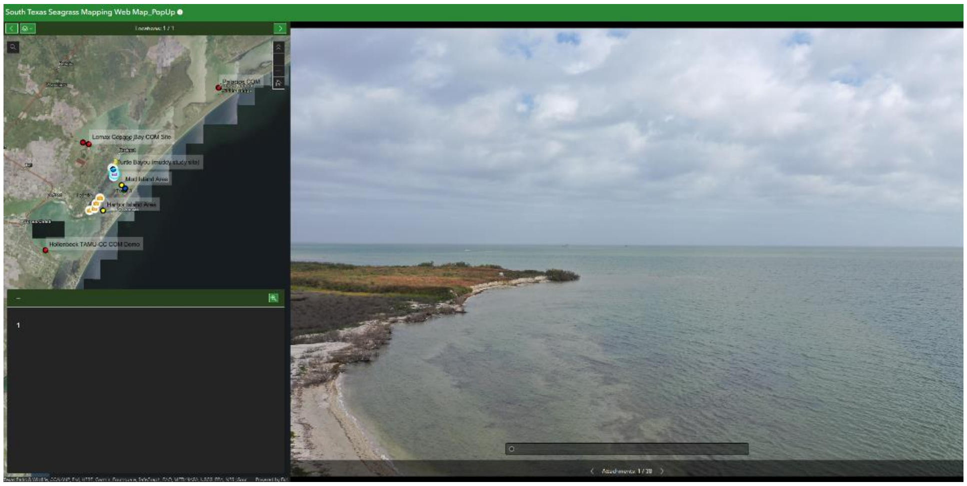
Original data will be freely shared and available to the scientific and general public upon request and via data repositories.

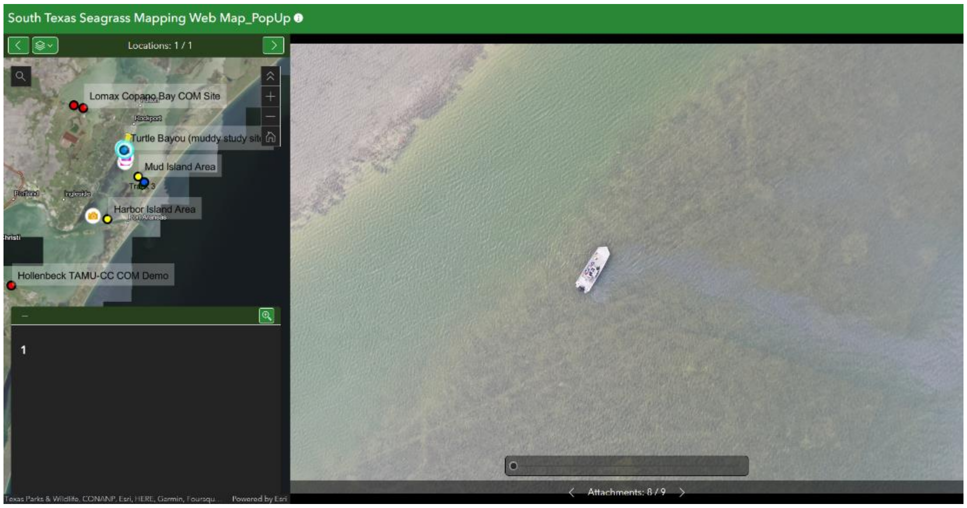
Website to share data

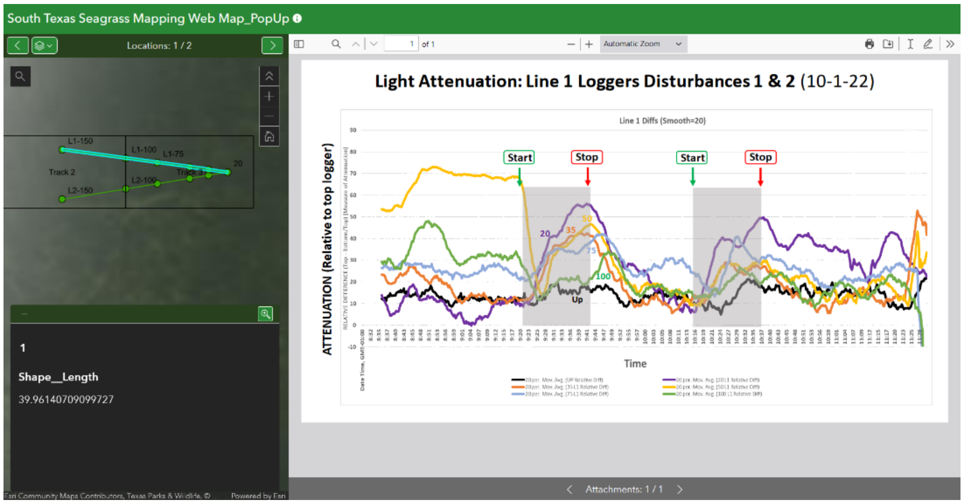
Dr. Zhang's "Integrated Information System for Oyster-Sediment-Seagrass Interactions" website

Project data will be available and linked to geospatial map information. Clicking on a map location will bring up some of the project's most informative data figures.
The current version of the website:
https://tamucc.maps.arcgis.com/apps/instant/attachmentviewer/index.html?appid=97b392e818294fb08c9390a389a0218d&locale=en.

Below are examples of some of the pages:






South Texas Seagrass Mapping Web Map_PopUp

Locations: 1 / 2

1

Shape__Length

39.96140709099727

Light Attenuation: Line 1 Loggers Disturbances 1 & 2 (10-1-22)

Attachments: 1 / 1


South Texas Seagrass Mapping Web Map_PopUp

Locations: 1 / 2

10-1 San Jose Dist #1 (9:21-9:41) ANNOTATED
Video # 0828 at **9:49 AM (8 min post-Dist #1)** [Collecting TSS]

10-1 San Jose Dist #1 (9:21-9:41)
Video # 0828 at 9:49 AM (8 min post-Dist #1) [Collecting TSS]

Attachments: 2 / 2

Cyverse Bisque

Cyverse has changed their operating procedures and can no longer provide free storage for all of our data as originally planned. However, a curated subset of imaging data (sediment plumes and seagrass leaves) will be prepared and uploaded to Cyverse Data Commons to be publicly available for analyses via Bisque (https://cyverse.org/bisque-image-analysis). This will include robust metadata and links to associated measurement data stored on the "Integrated Information System for Oyster-Sediment-Seagrass Interactions" website, or DRYAD for published work.

DRYAD

DRYAD (https://datadryad.org/stash) offers a permanent and stable repository, including a DOI, for data associated with peer-reviewed publications. Project images and data for our subsequent publication will be submitted there for storage and open accessibility.

Individual researcher's laboratory websites will contain data or links to other stores of project data.

Presentations & Publications

Both presentations and peer-reviewed publications will be used to disseminate project research findings:

Mehrube Mehrubeoglu*, **Kirk Cammarata**, **Hua Zhang**, Justin Giessel, and Lifford McLauchlan. “Sediment Plume and Seagrass Detection in Remote Sensing Imagery of Seagrass Beds” **Proceedings of The 26th International Conference on Image Processing, Computer Vision, & Pattern Recognition.** IPCV '22: July 25-28, 2022, USA

Mehrubeoglu: **Mehrube Mehrubeoglu, Kirk Cammarata, Hua Zhang**, Lifford McLauchlan “Plume motion characterization in UAV aerial video and imagery” 2023 SPIE Defense and Security, Conference 12528:Real-Time Image Processing and Deep Learning 2023, 30 April-4 May 2023, Orlando, FL. **Abstract Accepted**

J. Z. Giessel, M. Cortinas, T. Ha, "Segmentation of sediment plume using drone imagery" Final Class Project, EEEN 4333 Machine Vision and Image Processing Applications, Spring 2022 (Presented to class of 12 electrical engineering students).

Anticipated:

Cammarata et al:

Presentation at ASLO Ocean Sciences Meeting, Feb 2024 New Orleans, LA

Peer reviewed publication, probably PLOS ONE or Frontiers in Marine Science

TX Seagrass Monitoring Working Group

This project has been announced, along with associated updates, at the last two meetings of the TX Seagrass Monitoring Working Group. A presentation of the project’s findings is anticipated at the next meeting in late spring or summer. Feedback and suggestions will be solicited from members and added to this final report as an addendum.

Student Benefits

Three MS graduate students worked on the project as well as three undergraduates. One student’s Thesis and another’s Non-Thesis Project will be based on this project. These students are expected to present their work in various student research venues.

In addition, a class project in Dr. Mehrubeoglu’s Machine Vision and Image Processing class utilized project image data to learn analytical techniques. They presented their work to the class, which benefitted 18 students.

Stakeholder Input

Finalized copies of the Final Project Report will be disseminated to the following stakeholders seeking questions, comments and suggestions. Any input received will be shared in the addendum to the final report.

TPWD and MANERR colleagues, COM and seagrass researchers, commercial COM operators and members of the TX Seagrass Monitoring Working Group

SUMMARY AND CONCLUSIONS

This project set out to measure potential sediment generation at cultivated oyster mariculture (COM) operations in TX, then apply that knowledge to simulate sediment plumes with similar characteristics at test seagrass beds, to measure if there were impacts to the seagrass. It should be noted that “sediment plumes” or “sediments” are used here in the most general sense of suspended particles of inorganic or biotic nature that can settle or attenuate light penetration into the water column, regardless of source (NOAA). Observations and measurements were made at 3 small shallow water (wadable) research COM operations, and Texas’ first approved commercial scale suspended cage COM farm, which was located in relatively deeper water (8 ft). Operations and measurements were different in the 2 types of sites due to depth. Sediment plumes were characterized by aerial imagery, and measures of light attenuation, Total Suspended Solids (TSS) and settleable solids in sediment traps. At the same time, 2 seagrass study sites were selected based on large size, even-depth monotypic stands of *Halodule wrightii* in either sandy or muddy sediments. Two sites were selected and prepared for a BACI experimental design (Before-After and Control-Impacted) which required simulated sediment plume generation at a baseline location for Control vs Light vs Heavy in Tracks 1 - 3 respectively. These sediment disturbances were created by people walking back and forth along the disturbance baseline for specified times, standardized as person-minutes (p-m). Sediment plume characteristics and seagrass indicators (sediment grain size, leaf density, leaf length, biomass, belowground to aboveground biomass ratio, epiphyte to seagrass biomass ratio, and red to green epiphyte fluorescence ratio) were measured at different distances from the sediment plume source (50, 100 or 200 ft for Zones A – C respectively). The muddy seagrass study site had to be abandoned due to unacceptable hydrology. For the remaining sandy seagrass study site, sediment disturbances and measurements were made at different distances to test how far sediment plumes might travel and impact seagrasses, and over the course of a 5 month period (July – November) to record any cumulative effects and seasonal changes. The heavy disturbance Track 3 received 811 p-m compared to 455 p-m and 30 p-m respectively, for Tracks 2 and 1.

Summary of Findings from Each Area

Results from the principal measurements used to characterize sediment plumes at all study sites were summarized in the Summary Table at the end of the Results section. Additional figures there illustrate results from collective analyses of data from each of the shallow COM, deep COM and seagrass study sites.

Work at the **shallow research COM sites** included a small number of direct observations of operators performing typical tasks (cage flipping, bag shuffling and harvesting). However, at the time of this study, the Copano site was recently decommissioned, the Palacios site was winding operations down and getting smaller (ended in August 2022), and the Hollenbeck site (< 15 cages) was relatively new and growing in size. Aerial imagery could only be obtained at Palacios. Simulated sediment disturbances (people walking around the cage line) were performed 2-3 times (with increasing intensity) per each trip to these shallow COMs to perform necessary measurements. These high energy sites had high ambient turbidity and currents that shifted over time with wind and tides. Ideally, a line of poles would be placed down-current from the cageline at various distances, but getting alignment proved difficult. Multiple tracks of loggers were deployed to improve chances of capturing any sediment plumes. Most commonly, tracks of loggers had to be aligned somewhat perpendicular to the cageline, meaning any impacts derived from a small number of cages as opposed to if currents and measuring stations had all been parallel. Changes in relative light attenuation (at seagrass canopy height, but no actual seagrass) associated with disturbance were typically 5-20% and pulsating with each pass of a disturbance walker. Plumes were documented to travel at least 100 ft, but typically became indiscernable from background beyond that distance. By aerial imagery at Palacios, plumes became indiscernable from background turbidity after about 2 min. Sediment trap values represent sediment dry weight that was captured during the experimental deployment period (typically ~ 3-4 hrs) at the upstream control location or at lines of measuring stations down-current at various distances from the sediment plume source. Changing depths around the cages complicated ambient turbidity at Palacios and Hollenbeck. TSS measurements, retrieved by grab sampling immediately following a disturbance, were problematic due to timing of the sampler relative to the passing sediment plume, and to potential contamination issues when walking towards the sampling stations that were up-current of others.

Measurements at the **deep COM site** required deployment of measuring stations as floating cages where the sediment traps were typically ~ 4ft deep (site depth ~ 8 ft) and light attenuation could be measured at 2-4 ft depth. TSS samples were acquired from the surface. The water in Copano Bay is generally turbid, but it could vary greatly at the COM site depending on wind speed and direction. At times, the water was calm and relatively clear. Due to lack of bottom disturbance here, sediments mostly arose from operational manipulations of the oyster bags and cages, which shakes loose colonized biota. Aerial videography and image analysis showed that these mini sediment plumes were short-lived and small in area on relatively calm days. Measured values of light attenuation, TSS and settled sediments were typically smaller than at the shallow COM sites, but it was difficult to detect any features that can be definitively associated with a cage disturbance, even with measurement stations within 50 ft of a working area. Alignment with currents was again an issue. We typically deployed measuring stations 20-50 ft from cages or site boundaries at cardinal points (N-S & E-W) such that one station could, ideally, be defined as up-current and the other down-current. These could vary by day or even within a day. So values for “Differences *potentially* associated with activity” derive from the difference between the Up- and down-current stations, and presumably indicate what exits the farmed area from the surface to 4 ft deep. A direct plume-grab for TSS at the source cage showed 35 mg/L dry wt of solids as the peak value of a plume, and it had ~60% organic matter composition. These values were about on par with the highest observed measuring station values on a windy day. However, due to the changing conditions and uncertainties about hydrology and disturbed areas and timing, we cannot associate any values to COM operation activities directly. Any differences in values might represent a *maximum* since it would be difficult the parse the sources between ambient turbidity and farming activity. The data collected is summarized by range, mean and median values as guidelines for a starting point to understand these sites. UAS imagery was used to detect small cage-maintenance plumes, but these were short lived and small in area. The plumes dissipated without traveling very far so it is uncertain if an instrument could pick it up even if optimally positioned. An understanding of the hydrology of such sites is important for both performing measurements and for actually determining what, if any, potential sediment plume impacts could arise.

We did, however pick up some patterns and consistencies. Measures of light attenuation, TSS and sediment trap collections were all higher at the shallow COM sites compared to the deep COM site. Within the deep COM site, North had the lowest sediment collection, but that sediment collection had the greatest level of % organic composition compared to the South and West locations. The East sediment trap measurements presented wide variability with mean values near those of South and West (~2 fold lower), and a large discrepancy between mean and median values. Conversely, the % organic matter of East more closely resembled that of North.

Application of simulated sediment plumes at the **seagrass study sites** attempted to produce a range of sediment plume intensities and duration across different tracks and zones of the study sites. The Turtle Bayou muddy seagrass site had to be abandoned because the hydrology of the site was inconsistent with the experimental design and there was no workable adaptation. Hydrology and changing currents also presented difficulties at the San Jose sandy seagrass study site, but experimental design adaptations were used to achieve a reasonable facsimile of the original design to be able to compare the heaviest sediment impact at the 0-50 ft zone of Track 3 (S3-50) to the light disturbance zone of Track 2 (S2-50) in relation to the undisturbed control zones (S1-50) and upstream (S1-3 Up) control locations. In addition, comparisons could be made with distance away from the disturbance source (baseline) along each track as sediment plumes would be expected to dissipate with distance. Aerial imagery illustrated the consequences of unexpected currents and experimental adaptations, with a full analysis presented for the 10-1-22 disturbance set of measurements. In this case, the sediment plume travelled over 300 ft (perpendicular to the track orientation) at an average rate of about 10 ft/sec. This experimental orientation also ordinated applied disturbance (heavy – light – lightest) across Zone A of Tracks 1 – 3, respectively. Analyses of aerial imagery and light logger data provided similar estimates of sediment plume movements. We demonstrated gradients of light attenuation that coincided with sediment trap collections. Measures of applied seagrass sediment plumes suggest that their intensity characteristics were slightly greater than those observed at the deep suspended cage COM, but also slightly less than the observations at the shallow COMs. Different experiments may have had greater or smaller impacts individually.

Before (Pre), during (Mid) and after (Post) the seagrass site sediment plume disturbance experiments, we measured a **suite of seagrass indicators** to determine if there were any negative impacts of the sediment plumes. The seagrass indicators included % coverage by seagrass, leaf density, leaf length, biomass, belowground to aboveground biomass ratio, epiphyte to seagrass biomass ratio, epiphyte community fluorescence, epiphyte areal colonization of seagrass leaves, and invertebrate abundance and diversity. Comparisons of data were made across the 3 Tracks (sediment treatments), along the tracks between different distances from the disturbance source, across times (Pre, Mid, Post), and specifically between Track 3 Zone A and Track 1 Zone A (undisturbed) or Upstream sites. For the most part, we did not detect any significant differences in seagrass indicators between the heaviest seagrass disturbance zone (S3-50) and the lightest corresponding seagrass zone (S1-50) or the Upstream sites. This suggested no negative seagrass impacts. There was, however, a significant increase for invertebrate diversity at the most disturbed site (S3-50) relative to more distant locations in the same track (S3-200).

Overarching Conclusions and Discussion

The overarching conclusion from this short-term experimental work is that we found no evidence to suggest that a deep water suspended cage COM operation, with the limitations as currently permitted, results in suspended sediment levels that would impair seagrasses (*Halodule wrightii* specifically).

This applies to deeper water suspended cage COM where there is relatively little disturbance of the bottom. There is less certainty about potential impacts from the shallow COM operations as they tend to have higher ambient turbidity levels and their management would likely disturb bottom sediments on a regular basis. Nonetheless, the simulated plumes at the seagrass study site had similar light attenuation characteristics as those observed at the shallow COM sites and these experiments found no evidence of impaired seagrass over the short term. Hydrological conditions, distance from seagrass and farm size may be important in these situations. Although some sediment plumes were video documented from typical activity at both types of COM sites, we most consistently detected sediment disturbance parameters at the shallow sites. Plumes from individual cage handling at deep sites were not observed to travel far or persist for more than a few minutes. From our observations, bag shuffling to rearrange bags and/or take in for tumbling typically only occurred on a small number of cages (< 10) at 6 bags per cage, on a given day. Flipping might take place on a full row on a given day, but this level of activity would not cause a sediment plume of the magnitude that was generated at San Jose on 10-1-22. It is interesting to note that sediment plume water captured as a TSS sample directly adjacent to a flipped cage that had had bags shuffled showed similar % organic matter as an ambient water sample from a windy turbid water day (~50-60%).

Sedimentation simulations at the shallow seagrass site showed that tidal hydrology was the major driving factor for sediment plume travel. At times, intense plumes moved less than 50 ft, whereas at other times, they moved over 300 ft in 30 min (~10 ft/min). In addition, shipping activity from 5 miles away had significant impacts on sediment plume movements and seagrass plant orientation in seagrass beds. Current forcing from the shipping could override the ambient current conditions even on a moving tide. It should be noted that these observations applied to large oil tankers 5 miles away. In contrast we observed no effects from oil barges that passed about <1 mile away.

We applied 811 person-minutes of sediment disturbance to the heavy disturbance track of seagrass, mostly over a 4-month period. Yet on the background of natural seasonal and depth variations, no significant differences could be observed in comparison to seagrasses of similar depth at the undisturbed control track. This was true for a suite of seagrass indicators, including % coverage by seagrass, leaf density, leaf length, biomass, belowground to aboveground biomass ratio, epiphyte to seagrass biomass ratio, epiphyte fluorescence and invertebrate abundance and diversity.

An additional important conclusion was the indispensability of aerial imagery to this project. A great deal of information was obtained from imagery and, while it may not supplant direct measures of turbidity like TSS or light attenuation, it provides unique and instantaneous visualization of sediment plumes that would otherwise require extensive measurement work with a large number of spatially-arranged sampling stations. Aerial imagery allowed calculation of sediment plume movement rate and areal coverage and may potentially be useful in distinguishing sediment plume intensities and visualizing directional currents.

Limitations and Considerations

This conclusion applies specifically to deep water suspended cage COM operations and would not necessarily apply to shallow water situations where bottom sediments are repeatedly stirred up if they were in an area near pre-existing seagrasses. Our observations and measurements at COM sites and seagrass sites were limited due to a relatively short observation period, so the lack of observable sediment impacts on seagrasses cannot necessarily be extended to long term considerations (eg the 10 year permit period of COM operations in TX). At least one study associated seagrass impacts with accumulation of fine sediments (Zabarte-Maeztu et al 2020), so any activity producing large amounts of transportable fine-grained sediments is a potential concern. Our study did not analyze sediment plume particle sizes, but Labrie et al (2022) developed a spatial distribution model for the biodeposition of pseudofeces and feces for suspended cage culture based on tides, settling rates, wind and current velocity. They used detailed hydrological data for their site. The model was designed as a tool for managers to predict spatial deposition of organic matter in low energy environments. Such methods might be adaptable to develop a model for the high energy environments typical in Texas. But it’s clear that understanding potential travel of sediment/turbidity plumes requires knowledge of site-specific hydrology under varying weather conditions.

Another potential limitation in application of these conclusions is the difference in composition of the sediment plumes at COM sites (shallow vs deep) and those generated at the seagrass study site. Plumes from the latter presumably derived mostly from epiphytes and detritus. We observed that when a disturbance track occurred over seagrass, aerial imagery following disturbance showed bright green where the disturbance occurred, in contrast to the duller brown-green of the surrounding epiphytized seagrass. This shows that mobilized epiphytes contributed to the sediment plumes there. (Imagery shows still-intact vegetative cover at the disturbance track following at least 700 person-minutes of disturbance over 4 months.)

In contrast, sediments from the shallow COM sites in this study derived mostly from disturbed bottom sediments which would be a combination of native sediments in relatively high energy locations, plus any accumulated biodeposits. All of the shallow sites investigated were small-scale farms that were in the process of scaling-down or scaling-up, but not more than about 50 cages at the time of the study. Our study did not significantly represent cage-derived material at the shallow COM sites. The deep COM site investigated was deeper and required boat access. Routine farm activity did not appear to disturb bottom sediments, so all sediment plume particles derived from the suspended cages (on top of ambient water turbidity), which would be a combination of oyster biodeposits, colonized biofilms of algae and invertebrates, and ambient water column-derived particles entrained in the complex biofilm structure.

Comparison of the percent organic matter in TSS samples showed values ranging from 5% to 50% at Shallow COMs and 25% to 80% at the deep site (Mean values were 28% and 52%, respectively). For the seagrass disturbances, TSS % OM ranged from 20-50% for upstream and control areas but sediment disturbances showed a range from 5-73% (Mean values were ~30% in both cases). While increasing % OM trends with distance from the disturbance were expected, and sometimes observed, analysis of the data overall did not support this idea statistically. It should be noted that a number of TSS % OM samples were lost to sample processing errors and equipment failure in the laboratory, so data for seagrass TSS % OM is less robust.

Comparison of the percent organic matter in settled sediment trap samples showed values ranging from 0.3% to 39% at shallow COMs and 5% to 46% at the deep site (Mean values 2-3% and 9%, respectively). For the seagrass disturbances, settled sediments % OM ranged from 0.3-82% (mean 11%). Seagrass site sediment plumes thus had sediment trap % OM values more similar to the deep COM values, but TSS % OM values more similar to those observed at the shallow COM sites

To put these comparisons into perspective, it should be noted that TSS collections represent surface water, whereas sediment trap collections represent water near the bottom at seagrass canopy height. The difference was very small at the shallow seagrass site, reflecting somewhat similar values for % OM from TSS and sediment trap samples. However, the depth difference and % OM difference for the two measurements was much larger at the COM sites, which presumably reflects a bias in the particles assayed by each type of measure. The deep COM site was also different from the shallow COM site in that the sediment trap was suspended in the water column at just over 1 m depth, making it off the bottom but as deep in the water column as for the shallow sites. The range of % OM was greatest at the deep sites and the observation that the highest recorded values of TSS % OM occurred in some samples there is explained by a deficiency of suspended more-dense sandy sediments.

In consideration of the foregoing, the potential effects of simulated sediment plumes at the seagrass site may not entirely represent the potential effects of COM sediment plumes on seagrass, since the latter would likely contain pseudofeces and feces, and thus provide a different nutrient profile compared to that from seagrass sediment plumes. Oyster interactions with seagrass are complicated in natural systems, with both positive and negative interactions (Booth and Heck 2009; Plutchak et al 2010; Tallis et al 2009; Murata et al 2021; Huang et al 2023). The relationship may be density dependent in aquaculture associations (Booth and Heck 2009). Burial of seagrass by large amounts of sediment is another concern. Much of what is known regarding sediment physical impacts on seagrass comes from studies on impacts from dredging boat channels (Erftemeijer et al 2006), which are intense but short-lived, and are often regarded as temporary but recoverable. This type of sediment disturbance would probably differ from those of COM operations in many respects. In addition, Texas COM sites may not respond in the same ways as found for other studies because of the generally high energy conditions in TX waters.

Future Studies

To continue to fill important knowledge gaps, studies should accommodate longer term monitoring and reassessment. It is common for impacts to only manifest after years of cumulative effects. Methodology developed by Labrie et al (2022), making use of accurate, continuous ADCP current profiling, as well as sediment analyses, would provide a spatial understanding of hydrology and sediment accumulation. Likewise, understanding if seagrasses are susceptible to damage also requires some knowledge of the hydrology of a site.

Aerial imagery and subsequent quantitative analyses were important factors in this study and would also be of great benefit in future related work.

Another knowledge gap to consider is understanding the potential for impact from *in situ* cage cleaning. The potential economic benefit is clear, but little is known about what, if any, impacts might occur. Hydrology would certainly play a key role again. The potential for plumes of organic material is undeniable, but this could be readily tracked by aerial imagery as a sort of hydrological tracer, and that knowledge could guide studies of potential for impacts. It should be noted that the nutrients contained in oyster biodeposits and cage biofilms released by cage-cleaning, would be nutrients that were already in present in the water column and taken up by the respective organisms and released in concentrated form. In a sense, the COM is mainly concentrating and redistributing these nutrient resources in a localized trophic shuffle. Such knowledge might inform exploration of algal co-culture opportunities.

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