APPENDIX C. BENTHIC STUDY

Changes in Benthic Communities at Commercial Aggregate Mining Leases in Central San Francisco Bay

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Abstract
Sand mining in Central San Francisco Bay has been commercially important for more than 70 years. Over 1 million cubic yards of sandy sediments are extracted annually for concrete and fill material. San Francisco Bay also serves as an important fishery and critical habitat resource for >500 species of fishes and mammals, including special status species, such as Pacific herring, Pacific salmon, and Northern anchovy. Macrobenthic invertebrates additionally represent a key food source and biological indicator for the health of the Bay. Due to concerns for prolonged effects of sand mining activities on Bay habitat resources, this study aimed to evaluate the potential changes in benthic macroinvertebrate taxa and their physical habitat over a nine-year period using existing infauna data. Three benthic surveys that sampled sand mining leases in Central San Francisco Bay between 2008 and 2017 were the subject of the current work. A 2008 study sampled multiple lease areas in Central Bay, while the two latter studies in 2016 and 2017 focused on a single lease area, with each study also examining adjacent, unmined, control areas. The results of our evaluation highlighted strong spatial variation in benthic resources of Central Bay, that did not correlate with sand mining activities. The Central Bay infaunal community was predominantly represented by polychaete worms, nematodes, bivalve mollusks, and amphipod crustaceans, which seem to be adapted to the dynamic environment. Treatment type (mined vs. unmined) was not a significant driver of the non-metric multidimensional scaling or multivariate generalized linear models. Throughout the time series average abundance and diversity declined and was consistent between mined and unmined control areas. Time had an effect on total abundance and benthic diversity metrics, where the abundance of several taxa was associated with changes in water quality and sediment characteristics. The results support previous studies that have shown that natural perturbations of benthic habitat outweigh the potential short term effects following sand mining events, due to rapid recolonization of sediments. However, due to the focus on sampling in the low diversity estuarine sand assemblage of Central Bay in the two latter studies, there remains a need for information on current habitat conditions in polyhaline sand mining areas of San Francisco Bay to better understand long-term changes in benthic responses.

Highlight: Spatial variation in habitat drives benthic community patterns in sandy sediments of Central San Francisco Bay

Keywords: sand mining; benthic; macroinvertebrate; trends; variability; multivariate

Introduction

The mining of sand resources has the potential to impact native species that utilize such areas and the benthic resources they provide. Within San Francisco Bay (Bay), dredging of marine sediments is routinely conducted for the creation and maintenance of harbors, deepening of shipping channels, and for use as commercial aggregate. Dredging for harbors and shipping channels has been conducted in San Francisco Bay since the 1800s, whereas the dredging of sand for commercial construction activities (sand mining) has only been conducted since the 1930s (Hanson Environmental, 2004).

In Central San Francisco Bay, sand has been commercially dredged since the 1950s (Hanson Environmental, 2004) for use as construction fill material and making concrete. Currently, sand mining within the Bay only occurs within defined lease locations of Central Bay, Middle Ground Shoal, and along Suisun Bay channels (**Figure 1**). Approximately 1.6 million cubic yards of material are extracted per year by several marine aggregate companies that operate in the Bay. State and Federal permits currently allow for up to 2.1 million cubic yards to be extracted annually (Hanson Environmental, 2004).

Due to concerns about the potential effects of habitat disturbance on benthic biological communities in the Bay resulting from commercial aggregate mining, and a lack of applicable scientific studies concerning the subject, benthic surveys have been conducted to evaluate the effects of sand mining on these biological resources. Previously, three surveys (completed in 2008, 2016, and 2017) analyzed benthic communities at Central Bay sites and at Suisun Bay sites. Due to confounding sediment characteristics of the Suisun Bay sites among the three survey years (See **Appendix A1**), the present analysis will only use data from the Central Bay sites including control and mined areas.

To assess changes to benthic communities inhabiting sand mining areas in Central Bay, this study aimed to answer two key questions:

1. Are there statistically significant differences over time (i.e., nine years) in benthic communities inhabiting sand mining leases compared to unmined control sites in Central San Francisco Bay?
2. Are the potential changes over time in benthic communities at sand mining leases associated with environmental differences that are not observed at unmined control sites?

Methods

Study Area

The focus area of the current study is San Francisco Bay, located in northern California, USA. San Francisco Bay is comprised four sub-embayments (**Figure 1**), including (in order from south to north) South Bay, Central Bay, San Pablo Bay and Suisun Bay. The eastern portion of Suisun Bay connects with the Sacramento-San Joaquin Delta. The Golden Gate at the mouth of San Francisco Bay is the interface between the Central Bay and the open ocean. The channel floor under the Golden Gate is mainly comprised of bedrock with a maximum depth of 113 m and tidal currents that can peak at 2.5 m/s (Barnard et al., 2013). East of the Golden Gate, the deepest areas of the Central Bay have the coarsest sediments and strongest currents (Chin et al., 2010). These strong tidal currents flush mud and fine sediments out of the Central Bay, leaving behind coarser sediment than are present in the other embayments, which help to maintain the deeper water depths (NOAA, 2007).

Current sand mining leases are held in west Central Bay (**Figure 2**), where mining has occurred within the deep-water channels, and the substrate is dominated by sandy bedforms with mostly coarse mobile sand intermixed with pebble, cobble, and gravel (AMS, 2009; Barnard et al., 2013; NOAA, 2007). The data analyzed in this study has been restricted to those sites within the mining lease areas of Central Bay. The eastern portions of Central Bay are mud-dominated and have little to no bottom features (Barnard et al., 2013; and references there-in). Tidal mud flats and shallow areas are limited to the perimeter of the Central Bay and cover a smaller spatial extent than shallow mud flats in North and South Bay (NOAA, 2007).

Applied Marine Sciences-2008

Field Sampling

The first benthic survey was conducted by Applied Marine Sciences (AMS) from August 19-22, 2008, and August 25-26, 2008, at 40 sites (**Figure 2**; **Table 1**). These sites included 20 within mining leases and five outside lease areas (control) within Central Bay and 15 sites (10 mining, five control) within Suisun Bay (AMS, 2009). Mined sample sites were identified using the end of dredge track lines of known mining events from Hanson Aggregate and then randomly selected within the lease areas. The five control sites were randomly located in two unmined lease areas. At each sample site, sediment samples were collected for benthic infauna, grain size, and total organic carbon (TOC). If pre-selected sites had either too fine or coarse sediment texture and/or was too deep to represent mining areas, new sites were identified by sampling the area within a 100-500 m radius until the sediment texture and depth criteria were met.

Sediment Collection

Sediment samples were collected using a 0.1 m^2 modified Van Veen grab. In the field, the grab was split into two approximately equal portions using a dividing plate inserted into the grab. The benthic infaunal community was assessed from one half of the Van Veen grab (0.05 m^2). The floating organisms observed were carefully removed to the 1.0 mm jar. The subsampled sediment was washed through a 2.0 mm screen to remove large bivalves, worms, gastropods, any other large benthic organisms as well as shell fragments and other debris. These organisms were placed into the 1.0 mm sample jar. The sample was gently washed and screened through nested 0.5- and 1.0-mm mesh bags prior to sorting. The mesh bags were washed through corresponding 0.5- and 1.0-mm sieves before being placed into appropriately labeled sample jars. The organisms were sorted into major phyla groups and identified to the lowest taxonomic level possible. Specimens were saved for reference and validation where required.

The other half of the Van Veen grab sample was used to collect the sediment grain size and TOC samples. Sediment samples for grain size were collected at each station in pre-cleaned plastic sample jars. Additionally, samples for TOC were collected and placed into pre-cleaned glass jars with Teflon lids. Sediment grain size and TOC samples were analyzed by Columbia Analytical (Kelso, WA).

NewFields-2016 and 2017

Field Sampling

Two benthic surveys were conducted by Marine Taxonomic Services (MTS) from October 13-15, 2016, and October 23-25, 2017, respectively, at 40 sample sites across Central Bay and Suisun Bay. Sediment was collected at 20 sample sites per area (*i.e.,* 12 mined, 8 control) for benthic infaunal community, grain size, and TOC. The surveys included sampling at two sand mining permitted lease areas (**Table 1**, **Figure 2**): Central San Francisco Bay (Point Knox Shoal, Lease # 7779W) and Suisun Bay (Lease # 7781S). Sampling locations were chosen based on camera penetration (indicative of substrate consolidation), apparent grain size (*i.e.,* silt, sand, or gravel), miscellaneous factors (*i.e.,* biota, shell debris), and spatial distribution from Sediment Profile Images (SPI). Much like the 2008 study, mined sites were chosen in consultation with Hanson Aggregate and Line Marine to cover areas that had been and would be mined in the future. Control sites were chosen as unmined areas that had similar habitat characteristics near/within the target lease areas.

Sediment Collection

Surface sediment samples for benthic community analysis were collected using a Ponar grab (0.05 m^2) sampler at each location. Samples were sieved with filtered seawater using 0.5- and 1.0-mm stainless steel mesh screens to sort and collect benthic infauna. The 1.0-mm samples were processed for organism identification and abundance, whereas those from the 0.5-mm were preserved and archived. Organisms were sorted into major phyla groups and identified to the lowest practical taxonomic level.

An additional sediment Ponar grab sample was collected at each of the sample sites (**Figure 2**; **Table 1**;) to assess TOC and grain size distribution. Sediment was removed from the Ponar grab, homogenized, and placed into pre-cleaned containers provided by the analytical laboratory and appropriately labeled. Grain size distribution and TOC analyses were conducted by Test America (Pleasanton, CA).

Data Assembly and Analysis

Benthic Community Composition

Benthic community data were integrated from the AMS and NewFields studies. To ensure comparability between studies, only species abundance from the 1.0 mm AMS samples were used for comparison to the 1.0 mm samples from the NewFields studies. All abundances represent the number of individuals per 0.05 m^2. Species counts for raw abundance per sample were compiled and species names were standardized/updated to reflect current taxonomic listings. Species were previously identified and are named at the lowest taxonomic level possible. Additionally, lease names were standardized across studies and control/mined treatment areas were designated based on sample site locations within or outside of lease/mined areas.

Sediment Parameters

Grain size data for all sample locations were compiled from both studies. For each sample site within the Central Bay, this included the percent of TOC, gravel, coarse sand, medium sand, fine sand, silt, and clay. A review of sediment characteristics and grain size between Central Bay and Suisun Bay revealed spatial differences in these parameters (**Appendix A1**). Therefore, benthic community samples from both studies were restricted to Central Bay for the new analysis to assess the potential changes in community composition and abundance.

Environmental Parameters

Climate and water quality data were gathered from a variety of sources for San Francisco Bay to supplement the collected sediment parameters (for grain size and TOC) to assess other covariates with change to benthic community metrics over time. ‘Dayflow’ is the daily average outflow from the Sacramento-San Joaquin Delta. This is referred to as the “net Delta outflow index” (NDOI) and is an estimate of the net difference between ebbing and flooding tidal flows at Chipps Island computed as a daily average (CDWR, 2023). Additionally, the bottom position of X2, the 2 ppt salinity isohaline, is used as a habitat indicator as it marks the turbidity maximum and peaks in abundance for select estuarine species (Jassby et al., 1995). X2 is measured from the Golden Gate Bridge and shifts position based on freshwater flow and saltwater intrusion during wet and dry years (CDWR, 2023). Daily estimates of NDOI were used to calculate monthly mean NDOI, and the daily position of X2 was used to calculate the monthly mean X2 position. Monthly measurements of water quality parameters from the United States Geological Survey’s (USGS) Water Quality of San Francisco Bay Research and Monitoring Program were used to further characterize physical and biological water properties within Central Bay (Stations 17, 18, and 20; Cloern & Schraga, 2016; Schraga & Cloern, 2017). These parameters include temperature, salinity, chlorophyll-a, dissolved oxygen (DO), and suspended particulate matter. Depth-averaged values of each parameter were calculated at each station within Central Bay close to the benthic sampling sites (Stations 17, 18, and 20), which were then averaged to calculate a Central Bay monthly mean. Monthly observed precipitation totals (as rainfall) were also collated from the California Nevada River Forecast Center as well as total rainfall for the corresponding water year (CNRFC, 2023).

To use these additional parameters at the appropriate timescale, monthly mean NDOI, position of X2 as well as water quality parameters were lagged three months. Total monthly precipitation for the same corresponding lagged months were used for rainfall totals. Therefore, the parameters associated with the AMS August 2008 benthic survey are averages from May 2008. Whereas parameters for the October 2016/2017 NewFields datasets are averages from July 2016 and July 2017. The three-month lag was used as an estimate of the response time of benthic community to climate perturbations, where selected parameters would shape species recruitment and distributions during the early pelagic lifecycle of benthic organisms (Widdows, 1991; Giangrande et al., 1994).

Diversity Metrics

All data and statistical analyses were conducted within the R framework (R-Studio Version 2022.12.0; R Core Team, 2022). Benthic community diversity metrics were calculated using the *diversity* function within the *vegan* community ecology package (Oksanen et al., 2022). Species diversity metrics, including species richness (*S*), Shannon's (*H*) diversity Index, and Simpson's (*D*) Index, are one of the most widely used metrics for assessing ecological communities (Magurran & McGill, 2011). Species richness, Shannon's index, and Simpson's index were calculated for each sample. Species richness (*S*) is the total number of species present within a community (Fisher et al., 1943). The Shannon's (*H*) diversity Index or Shannon-Weaver, Shannon-Weiner Index (Shannon, 1948) uses both the richness of species and their evenness or relative abundance. Similarly, the Simpson's (*D)* Index also uses richness and evenness but measures the probability that two randomly selected individuals will be from the same species (Simpson, 1949).

Analysis of Variance

Differences between treatment and timing were tested with a two-way ANOVA for total abundance, diversity metrics, species-specific abundance, and sediment parameters. Parameters were rank transformed to meet normality and equality of variance assumptions as needed. A Tukey HSD pairwise comparison was run post-hoc. For parameters that did not meet two-way ANOVA assumptions, differences between timing and treatment were tested using a nonparametric Aligned Rank Transformation analysis followed by pairwise contrast test post-hoc. Results of post-hoc tests of treatment x time effects are indicated by letter annotations on relevant figures where if two groups share a letter, it indicates that the p value for the pairwise comparison was not smaller than the specified alpha level (5%).

Multivariate Analysis

Two methods were used to test for differences in benthic communities among mined and control sites in Central San Francisco Bay. Firstly, non-metric multidimensional scaling (NMDS) ordination analysis was used to visualize differences in benthic community composition and the environmental variables, as well as to identify the species-specific drivers to focus on in the generalized linear models (GLM). NMDS is a rank-based approach which uses an indirect gradient that produces an ordination-based distance matrix, which represents the pairwise dissimilarity between objects in low-dimensional space (Kenkel & Orloci, 1986). Permutational analysis of variance (PERMANOVA) further partitions the dissimilarity matrix to compare variability between and within groups (Anderson, 2001, 2017). A species x site matrix of raw abundance was used in the NMDS ordination analysis. NMDS analysis was conducted using the *metaMDS* function from the *vegan* community ecology package, which transformed raw abundances using the Wisconsin double standardization and created a Bray-Curtis dissimilarity matrix (Oksanen et al., 2022). The ordination was performed from several random starts until a convergent solution was found. The effect of sediment grain-size as well as sample time, lease area, and treatment (control/mined) were determined using the *envfit* function. Additionally, species driving differences in benthic communities within Central Bay were identified with the *envfit* function as well. A PERMANOVA analysis was used to determine any significant differences between sample time periods, lease areas as well as control vs. mined areas using the *adonis* function in the *vegan* community ecology package with 999 permutations (Oksanen et al., 2022). Potential indicator species associated with time, lease, or treatment groups were identified using the *multipatt* function within the *indicspecies* package (De Cáceres & Legendre, 2009).

Following the NMDS ordination analysis, correlation and GLM tested for statistical significance of the environmental variables relative to benthic responses, focusing on the benthic metrics indicated by the NMDS and PERMANOVAs. Correlation statistics were used to select among the candidate environmental variables in relation to benthic community indicators. Subsequently, GLMs of benthic responses (i.e., species richness, abundance, and selected taxonomic groups) tested for the significance of categorical variables for treatment (mined vs. control) and lease, along with the candidate set of environmental parameters. The best and most parsimonious models to describe benthic responses were selected by corrected Akaike Information Criteria (AICc; Sakamoto et al., 1986) rankings, where all variables had a statistically significant parameter estimates at $p < 0.05$. The influence of outliers was assessed using the *car* package (Fox & Weisberg, 2019). Finally, the residuals of the statistical models were tested for the effects of time, by a one-way ANOVA test.

Results and Discussion

Are there statistically significant differences over time in benthic communities inhabiting sand mining leases compared to unmined control sites in Central San Francisco Bay?

Comparing the taxonomic composition between mined and unmined control areas among the three benthic studies revealed significantly higher total abundances in 2008 for both mined and unmined areas, compared to 2016 and 2017 ($p < 0.00001$). In 2008, mined leases had an average of >100 individuals compared to ~75 individuals from unmined control areas (**Figure 3**), which was not statistically different ($p = 0.99$). While in 2016 and 2017, the average total abundance was significantly lower with ~25 individuals. There was no statistical difference observed between mined and unmined areas ($p = 0.99$; $p = 0.56$, respectively).

Mean species richness was highest for mined and unmined areas in 2008 and 2016 compared to 2017 (**Figure 4**). Mined leases in 2017 had the lowest overall mean species richness. Species richness was significantly different between 2017 and 2008 ($p = 0.0002$), and 2017 and 2016 ($p = 0.011$), with no difference between mined and unmined areas ($p = 0.427$). There was no significant difference between mined and unmined areas for Shannon's or Simpson's indices ($p = 0.897$ and $p = 0.983$, respectively). Timing was not significant for Shannon's Index ($p = 0.105$)

or Simpson's Index ($p = 0.127$). Mean Shannon's and Simpson's indices were highest within the control leases of 2016, followed by the mined areas of 2008 and 2016 (Figure 5B, 5C). Overall, there were fewer species and total individuals in 2016 and 2017 than in the earlier study, which was consistent between mined and unmined areas. It is likely that the larger spatial coverage of the 2008 study included areas of relatively higher abundance and diversity. This hypothesis is further explored through the analyses that follow.

The taxonomic groups with the most abundant species across all three surveys included Amphipoda, Bivalvia, Gastropoda, Isopoda, Nematoda, and Polychaeta (**Figure 5**). These six taxonomic groups represented 72% (2016), 95% (2017), and 96% (2008) of the total abundance in each year. Generally, there was greater variability between sampling sites in abundance in 2008 compared to other years, reflected by the larger standard deviations. Nematoda species were notably the most abundant taxa in 2008, with similar abundances between mined and control samples, but were entirely absent in 2016 and 2017. Polychaetas dominated the benthic community in 2016 and 2017. Polychaete abundance was significantly different between 2008 - 2016 ($p = 0.04$), however there was no difference between mined and control areas ($p = 0.69$).Overall, polychaetes were the second most abundant taxa representative of the benthic community, with 32% of the total abundance in 2008, and > 50% in both 2016 and 2017. Amphipoda species were most prevalent in the mined areas in 2008 compared to control areas ($p = 0.002$) and were found in relatively low numbers in 2016 and 2017. Bivalve species were found in each sample every year, with the highest mean abundances in mined areas from 2008 ($p = 0.034$). Mean gastropod abundances were relatively similar among treatments years during 2016 and 2017 varying by 0-4 individuals. Mined areas were different from control in 2008 ($p = 0.006$). Notably, species within the Isopoda and Nematoda groups were only observed in 2008 samples, suggesting some potential differences from the 2016 and 2017 samples. Isopod abundances were also significantly different between mined and control areas in 2008 ($p = 0.0001$). Nematoda abundances in 2008 (mined and control) differed from both 2016 and 2017 ($p <0.0001$).

Benthic community analysis has previously identified five distinct benthic assemblages in the San Francisco Bay-Delta (Thompson et al. 2011), distributed along a salinity gradient: a polyhaline assemblage that includes Central Bay; a mesohaline assemblage that includes San Pablo and South Bay; an oligohaline assemblage comprised of Suisun Bay; a low diversity estuarine sand assemblage that occurs throughout the Bay-Delta system; and a tidal freshwater upper estuary assemblage (Thompson et al., 2011). Two of these five assemblages are located within or adjacent to the mining lease areas within Central Bay: the polyhaline assemblage and the low diversity estuarine sand assemblage. Thompson et al. (2011) found mean taxa richness for the polyhaline and estuary sandy assemblages to average 28 and 7 species, respectively, with ranges between 5 –59 and 1-17 species. Additionally, total abundance in polyhaline sites ranged from 8 – 12670, mean = 1694 and estuary sand sites ranged from 1-112 (mean = 36). Overall species abundances for both unmined and mined areas presented here fell within these reported ranges. A multiyear study of benthic macroinvertebrates of San Francisco Bay, observed that benthic populations often exhibit within-year fluctuations based on environmental perturbations and that the year-to-year predictability of species abundances was low. Yet, the overall assemblage of benthic species is more stable over longer time scales (Nichols & Thompson 1985).

The species assemblage of the estuary sand and polyhaline assemblages described by Thompson et al. (2011) are encompassed in both the unmined and mined areas of this study. The estuary sand assemblage of San Francisco Bay was dominated by two polychaete species, *Heteropodarke heteromorpha* and *Hessionura coineau.* Low abundances of oligochaetes *Tubificidae;* amphipods *Grandifoxus grandis*, *Ampelisca abdita*, *Monocorophium acherusicum*, and *Corophium heteroceratum*; bivalves *Corbula amurensis* and *Tellina nucluoides*; polychaetes *Mediomastus* spp., *Streblospio benedicti*; and a single cumacean *Nippoleucon hinumensis* were also observed in estuary sand habitat (Thompson et al., 2011). Additionally, this benthic assemblage was categorized by low species abundance likely due to the sandy substrate and elevated levels of current disturbance. In contrast, Thompson et al. (2011) reported that the polyhaline assemblage was mostly comprised of several amphipods *A. abdita, M. acheruscium, C. heteroceratum, Photis brevipes, Corophium alienense, Americorophium stimpsoni,* and *Americorophium spinicorne;* oligochaetes *Tubificidae;* polychaetes *Mediomastus* spp., *Exogone lourei, H. heteromorpha,* and *S. benedicti;* cumacean *N. hinumensis;* and the bivalve *C. amurensis*. Samples from the polyhaline assemblage had the highest average number of taxa of the benthic assemblages. It also had the highest abundances across all assemblages due to large numbers of *M. acheruscium* (Thompson et al., 2011). Sediments within this assemblage were mostly silt and clay. It is evident that the surveys conducted in 2016 and 2017 are more characteristic of the estuarine sand assemblage of Central Bay, consisting of low diversity and high dominance of polychaete and amphipod taxa. The survey from 2008 exhibited a relatively higher number of species and total abundances in sand mining areas that are characteristic of the polyhaline assemblage (Ranasinghe et al. 2009) of San Francisco Bay. Overall, our analysis found that benthic communities inhabiting mined portions of sand mining leases have similar species assemblages compared to unmined control sites in Central San Francisco Bay, which includes those within this study and areas from Ranasinghe et al., 2009 and Thompson et al., 2011.

Are the potential changes over time in benthic communities at sand mining leases associated with environmental variables that are not observed at unmined control sites?

Sediment quality parameters presented the most direct method to assess environmental differences between mining areas (leases) and unmined control sites over time. While community abundances were lowest in 2016 and 2017, TOC was highest in 2017 compared to 2008 and 2016 (**Figure 6**, **Figure 7**).

Mean TOC in 2008 and 2016 was observed below 0.5%, whereas mean TOC in 2017 was up to 3x higher varying between ~1.25-1.5%. As such, TOC in 2017 was significantly different from

2008 and 2016 ($p < 0.0001$). However, there was no difference between mined and unmined areas ($p = 0.52$). The observed TOC values in mined and control areas fall within previously reported ranges for polyhaline and estuary sand habitat in Central Bay (0.1-3.77%, mean = 1.14% and 0.08-2.7%, mean = 0.52, respectively; Thompson et al., 2011). Peak mean species richness has been recorded at TOC concentrations between 0.5-1% (converted mg/g to %; Hyland et al., 2005). For sampling sites across all years, when TOC was low (0-1%), these areas had the highest animal abundance (>100 individuals; **Figure 7**). In contrast, sampling sites in 2016/2017, which have higher TOC contributions (>2%), had lower animal abundance (1-50 individuals; **Figure 7**). Areas with low to moderate TOC concentrations and low species richness are known to occur due to factors such as natural salinity and grainsize gradients, insufficient organic matter input, and natural disturbance such as erosion from strong currents, or other geochemical, physical, or hydrodynamic conditions (Hyland et al., 2005).

Differences between mined and unmined areas in the percentages of coarse sand, fine sand, silt, and clay varied by year (**Figure 6**). Mean percentages of gravel and medium sand were consistently greater in mined and control sites, respectively, in each year. Average gravel contributions were mostly <10% in control areas but were relatively high for mined areas in 2008 and 2017. However, there were no significant effects of treatment or timing on gravel percentages ($p = 0.56$; $p = 0.13$, respectively). Highest mean coarse sand was also found in mined areas in 2008 but was under 20% in the other years. Mean medium sand contributions were higher in unmined areas than in mined areas for 2008 and 2016 ($p = 0.007$). Fine sand was consistent across years, although mined areas in 2016 had the highest mean percentage (>40%). For both coarse and fine sand percentages, there were no significant differences between treatment and timing (coarse: $p = 0.20$, $p = 0.37$; fine sand: $p = 0.60$, $p = 0.26$, respectively). The mean percentage of silt was notable for decreasing over time in mined areas, where mined areas in 2008 had the highest overall silt contribution (~1.75%) but this was not significant ($p = 0.09$). Mean silt percentage was slightly higher in control sites in 2016, relative to control sites in 2008 and 2017, which was also not significant ($p = 0.30$). Similarly, the contribution of clay to the sediment pool was highest (~1.5%) in the mined areas from 2008 and 2016 and lowest in both control and mined areas in 2017. As such, there were significant differences in treatment ($p = 0.047$) and timing ($p < 0.0001$) for percent clay between 2017 and both 2008 and 2016.

Benthic assemblages from the three benthic studies most strongly grouped by time, and then by lease (**Figure 8**, stress = 0.1432). Out of 23 taxa identified as potential community drivers of the NMDS ordination, 14 were found to be significant (**Figure 8, Table 2**). Of the seven sediment parameters, only percent clay was significant in differentiating benthic communities ($r^2 = 0.104$, p-value = 0.038, **Figure 8B, Table 3**), however percent TOC and percent silt were only slightly non-significant ($r^2 = 0.085$ and 0.084, p-value = 0.065 and 0.056, respectively). The time-period of sampling along with lease were significant drivers of benthic communities (r^2 = 0.61 and 0.56, p-value = 0.001 and 0.001, **Figure 8B, Table 3**). PERMANOVA results (**Table 4**) confirm community differences based on time ($r^2 = 0.21$, p-value = 0.001), however lease was not significant (r^2 = 0.16, p-value = 0.10). Treatment (control vs. mined) was also found to be not significant from the PERMANOVA analysis (r^2 = 0.011, p-value = 0.48).

The sediment parameters of benthic habitats in sand mining areas within Central Bay appear to vary substantially among locations. Mean abundance of 13 taxa identified as significant drivers of the NMDS ordination was binned according to four grain size categories that span the range from coarse grains (>20% coarse sand) to very fine sediment (>1.5% Clay) (**Figure 9**). Several of the taxa revealed distinct associations among sand mining areas. The polychaete taxon *Glycinde* spp. exhibited higher abundances in mined leases with higher percentages of silt and clay relative to sand mining areas with coarser sand grains. Conversely, the polychaetes *Heteropodarke heteromorpha* and *Nematoda* spp. exhibited strong disassociations with fine sediments in mining areas, occurring predominantly in coarse and medium sand. Medium sand contributions in the North Sea dictated functional characteristics of benthic communities in aggregate mined areas (Goedefroo et al., 2023). Polychaetes are often used as indicators of environmental disturbance exhibiting both sensitive and tolerant taxa (Pocklington & Wells, 1992). Opportunistic species, such as *Capitella capitata* (Tsutsumi, 1987), are known to recolonize disturbed sediments quickly as a feature of their reproductive strategy. *Sacrocirrus* spp., which is one of the polychaete taxa that did not strongly differ by grain size, are known as “disturbance specialists” that exhibit behavioral traits to tolerate highly dynamic environments (Glasby et al., 2021). Areas that experience periodic maintenance dredging in Central Bay exhibit similar species composition, where macroinvertebrate densities are typically dominated by polychaetes and amphipods but also include oligochaetes, nematodes, and bivalves (De La Cruz, 2020). Dredged areas close to sand mining leases were dominated by polychaetes, nematodes, and oligochaetes in 2013, where un-dredged areas were dominated by nematodes, polychaetes, amphipods, and oligochaetes (De La Cruz, 2020). In 2014, dredged areas near Central Bay sand mining lease areas were dominated by polychaetes, oligochaetes, and nematodes, whereas un-dredged areas included polychaetes, oligochaetes, and amphipods (De La Cruz, 2020).

Benthic community responses in Central San Francisco Bay thus exhibit broad scale spatial variability in habitat conditions that do not appear to significantly differ between mining leases and unmined control areas. Generalized linear models indicated that factors associated with habitat variability predominate over other potential factors (**Table 5**). The best statistical models of benthic response were composed of general water quality or habitat variables, rather than a treatment effect. Species richness, total abundance, and several species-specific abundances were parameters that differed among years. While it is unlikely that the specific water quality indicators were associated with the observed changes, the significant association on total abundance and richness suggests difference in environmental conditions influences the community patterns among the various sites sampled. Finally, lease was a significant factor in three of the statistical models, further supporting the observation that spatial variability is a dominant feature structuring the benthic communities of Central Bay.

Soft bottom benthic communities within Central Bay and San Francisco Bay are known to vary with water depth, salinity, and sediment grain size (Nichols & Thompson, 1985; Thompson et al., 2011). The sand mining lease areas within Central Bay are in areas with fluctuating salinity exposed to physical disturbance due to high currents that can produce large sand wave fields. Infaunal distribution and abundance correlated best with the relative percentages of clay and gravel in sediments, while the dominant species groups in Central Bay were indicative of disturbance whether within or outside mining leases. Previous monitoring of the direct effects of sand mining has shown that benthic species can recolonize quickly (e.g., Constantino et al., 2009). Rates of recovery of six to eight months have been shown to characterize many estuarine sediment communities where frequent disturbance of the habitat occurs (Newell et al., 1998). In addition to naturally varying parameters such as water depth and salinity, other physical environmental differences between lease areas may have affected infaunal community patterns. Impacts to the benthic community are expected from physical removal of sediments and infauna. Based on previous studies, recovery of species composition may be expected to take several years to occur, as a function of hydrodynamics, substrate characteristics, chemicals, and biotic interactions (Snelgrove et al., 1993; Sundberg and Kennedy, 1993; Ólfasson et al., 1994; Hsieh & Hsu, 1999; Snelgrove et al., 1996). These results suggest duration of benthic effects may differ with location of mined sites, due to physical and biological differences across the region (Nichols & Thompson, 1985; Hyland et al., 2005; Thompson et al., 2011).

Conclusions

Benthic studies aimed at identifying changes in benthic community dynamics as a direct result of sand mining require study designs that sample the extent of natural spatial variability. The inferences from the three Central Bay benthic surveys evaluated here are that the variability in benthic community distributions in response to habitat and natural perturbations far outweighs any potential affect from sand mining disturbances. Thus, concerns for the prolonged effects of sand mining activities on habitat resources for higher trophic level species that inhabit Central San Francisco Bay are not supported by these results. However, inter-annual variability also clearly distinguished the benthic conditions during the 2008 study from the more recent studies in 2016 and 2017. This appears to be largely a function of sampling of the estuarine sandy assemblage in the latter studies compared to sampling of both the estuarine sand and polyhaline assemblage in the earlier study. Benthic community abundances in San Francisco Bay strongly associate with salinity gradients, which likely explains the larger variation seen in the 2008 study and relatively lower diversity of species encountered during the latter period. Consequently, the spatio-temporal evaluations conducted for our study lack detailed investigation of the impacts of sand mining on the polyhaline benthic assemblage over time. Thus, a future comparative study that samples in multiple sand mining leases and unmined sites of Central Bay encompassing the benthic assemblages discussed here would provide the needed information regarding benthic recovery and the prolonged effects of direct sand mining activity on the benthic macroinvertebrate community. Information from the comparative surveys could also be used to substantiate the lower quantification of benthic species abundance and diversity within the estuarine sandy habitats from the recent studies. It is necessary to sample sufficient spatial variation to quantify the rate of benthic macroinvertebrate recolonization of subtidal habitat disturbed by sand mining activity.

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Tables

Table 1. *Sample site coordinates for each benthic survey and their associated lease and treatment.*

Lease	Site Name	Year Sampled	Latitude	Longitude	Treatment	Group
PRC 2036	CB-2036-01	2008	37.840932	-122.449567	Mined	AMS
PRC 2036	CB-2036-02	2008	37.842329	-122.456073	Mined	AMS
PRC 709 East	CB-709E-01	2008	37.824633	-122.43545	Mined	AMS
PRC 709 East	CB-709E-02	2008	37.827773	-122.427607	Mined	AMS
PRC 709 North	CB-709N-01	2008	37.85305	-122.446517	Mined	AMS
PRC 709 North	CB-709N-02	2008	37.845217	-122.456017	Mined	AMS
PRC 709 North	CB-709N-03	2008	37.846267	-122.443083	Mined	AMS
PRC 709 South	CB-709S-01	2008	37.816212	-122.450662	Mined	AMS
PRC 709 South	CB-709S-02	2008	37.81436	-122.452449	Mined	AMS
PRC 709 South	CB-709S-03	2008	37.813333	-122.448258	Mined	AMS
PRC 7779 East	CB-7779E-01	2008	37.8459	-122.42815	Mined	AMS
PRC 7779 East	CB-7779E-02	2008	37.8463	-122.431	Mined	AMS
PRC 7779 North	CB-7779N-01	2008	37.859883	-122.450617	Mined	AMS
PRC 7779 North	CB-7779N-02	2008	37.858167	-122.453683	Mined	AMS
PRC 7779 West	CB-7779W-01	2008	37.832567	-122.437105	Mined	AMS
PRC 7779 West	CB-7779W-02	2008	37.836731	-122.453596	Mined	AMS
PRC 7779 West	CB-7779W-03	2008	37.8359	-122.452867	Mined	AMS
PRC 7779 West	CB-7779W-04	2008	37.831345	-122.459067	Mined	AMS
PRC 7780 North	CB-7780N-01	2008	37.8318	-122.429867	Mined	AMS
PRC 7780 South	CB-7780S-01	2008	37.820433	-122.4331	Mined	AMS
PRC 7780 South	CB-7780S-02	2008	37.816067	-122.429217	Mined	AMS
CB_Control_North	CB-CBCN-01	2008	37.849233	-122.437433	Control	AMS
CB_Control_North	CB-CBCN-02	2008	37.84505	-122.436067	Control	AMS
CB_Control_North	CB-CBCN-03	2008	37.8434	-122.434817	Control	AMS

CB_Control_South	CB-CBCS-04	2008	37.81755	-122.437609	Control	AMS
CB_Control_South	CB-CBCS-05	2008	37.814933	-122.437333	Control	AMS
PRC 7779 West	CB01	2016, 2017	37.832788	-122.437255	Mined	NewFields
PRC 7779 West	CB03	2016, 2017	37.832853	-122.435636	Mined	NewFields
PRC 7779 West	CB09	2016, 2017	37.832272	-122.436046	Mined	NewFields
PRC 7779 West	CB10	2016, 2017	37.83236	-122.435518	Mined	NewFields
PRC 7779 West	CB14	2016, 2017	37.831807	-122.437293	Mined	NewFields
PRC 7779 West	CB17	2016, 2017	37.831822	-122.435433	Mined	NewFields
PRC 7779 West	CB18	2016, 2017	37.831822	-122.43475	Mined	NewFields
PRC 7779 West	CB25	2016, 2017	37.831363	-122.434825	Mined	NewFields
PRC 7779 West	CB28	2016, 2017	37.830857	-122.437193	Mined	NewFields
PRC 7779 West	CB31	2016, 2017	37.830783	-122.435361	Mined	NewFields
PRC 7779 West	CB35	2016, 2017	37.830277	-122.43717	Mined	NewFields
PRC 7779 West	CB38	2016, 2017	37.830313	-122.434838	Mined	NewFields
PRC 7779 West	CBR03	2016, 2017	37.83383	-122.433678	Control	NewFields
PRC 7779 West	CBR04	2016, 2017	37.833505	-122.436971	Control	NewFields
PRC 7779 West	CBR07	2016, 2017	37.833155	-122.437551	Control	NewFields
PRC 7779 West	CBR09	2016, 2017	37.833163	-122.434853	Control	NewFields
CB_Control_PKS	CBR11	2016, 2017	37.829753	-122.437335	Control	NewFields
CB_Control_PKS	CBR14	2016, 2017	37.82982	-122.43347	Control	NewFields
CB_Control_PKS	CBR16	2016, 2017	37.82939	-122.435451	Control	NewFields
CB_Control_PKS	CBR19	2016, 2017	37.82911	-122.435986	Control	NewFields

Table 2. *Results from the species-specific Envfit analysis. Statistical significance (*) was assessed at p < 0.05.*

TAXONOMIC GROUP	SPECIES	NMDS1	NMDS2	r^2	P-Value
CHORDATA	*Branchiostoma californiense*	0.34752	0.93767	0.1209	**0.016***
GASTROPODA	*Callianax biplicata*	0.00473	0.99999	0.0900	0.055·
CNIDARIA	*Edwardsia* spp. *A*	0.22038	0.97541	0.0579	0.082·
POLYCHAETA	*Glycera oxycephala*	-0.65242	0.75786	0.1002	**0.025***
POLYCHAETA	*Glycera* spp.	-0.59471	0.80394	0.2870	**0.001*****
POLYCHAETA	*Glycera* spp. *A*	-0.97236	-0.23328	0.0775	0.068·
POLYCHAETA	*Glycinde picta*	-0.38845	-0.92147	0.0668	0.078·
POLYCHAETA	*Glycinde* spp.	-0.61009	-0.79233	0.1467	**0.008****
POLYCHAETA	*Hemipodia simplex*	0.67686	0.73611	0.4461	**0.001*****
POLYCHAETA	*Heteropodarke heteromorpha*	-0.75747	0.65287	0.2635	**0.001*****
ISOPODA	*Ianiropsis* spp. *A*	-0.55336	0.83295	0.0597	0.061·
CUMACEA	*Lamprops quadriplicata*	-0.99998	0.00660	0.1726	**0.004****
POLYCHAETA	*Malmgreniella* spp.	-0.37306	-0.92781	0.0881	**0.044***
NEMERTEA	*Micrura* spp.	0.47145	-0.88189	0.1829	**0.003****
BIVALVIA	*Mytilidae* spp. *A*	-0.46652	-0.88451	0.0563	0.098·
NEMATODA	*Nematoda* spp.	-0.91026	0.41404	0.2787	**0.001*****
NEMERTEA	*Nemertea* spp.	-0.42846	-0.90356	0.0726	0.062·
POLYCHAETA	*Nepthys caecoides*	-0.07091	-0.99748	0.1303	**0.011***
BIVALVIA	*Nutricola confusa*	-0.46141	-0.88719	0.0602	0.069·
BIVALVIA	*Nutricola* spp.	-0.35160	-0.93615	0.1031	**0.025***
POLYCHAETA	*Opheliidae* spp.	-0.99428	0.10678	0.1039	**0.027***
BIVALVIA	*Rochefortia tumida*	-0.046652	-0.88451	0.0563	0.098·
BIVALVIA	*Tellina carpenteri*	0.99769	-0.06788	0.0885	0.059·

Table 3. *Results from the sediment characteristic specific Envfit analysis. Statistical significance was assessed at $p < 0.05$ (*) with highly significant p-values indicated by ***.*

VARIABLE	NMDS1	NMDS2	r^2	P-VALUE
TOC %	0.67504	-0.73778	0.0848	0.065·
GRAVEL %	0.03039	-0.99954	0.0253	0.458
COARSE SAND %	-0.96879	-0.24787	0.0157	0.604
MEDIUM SAND %	0.98041	-0.19697	0.0214	0.493
FINE SAND %	-0.22545	0.97425	0.0242	0.440
SILT %	-0.59534	-0.80347	0.0835	0.056·
CLAY %	-0.67091	0.74154	0.1042	**0.038***
TIMING			0.6100	**0.001*****
TREATMENT			0.0297	0.145
LEASE			0.5569	**0.001*****

Table 4. *PERMANOVA results for differences in benthic communities based on timing, treatment, and lease. Statistical significance was assessed at $p < 0.05$ (*) with highly significant p-values indicated by ***.*

VARIABLE	DF	SUM OF SQUARES	r^2	F	P-VALUE
TIMING	2	5.5853	0.20990	8.4412	**0.001*****
LEASE	11	4.1639	0.15648	1.1442	0.100·
TREATMENT	1	0.3187	0.01198	0.9634	0.476
RESIDUAL	50	16.5418	0.62164		
TOTAL	64	26.0698	1.00000		

***Table 5**. The highest ranked statistical models to test for the effects of climate, water quality, habitat, and time on abundance and diversity of benthic indicators in Central San Francisco Bay. All models consisting of climate and water quality models were statistically significant at alpha = 0.05.*

	EFFECTS OF CLIMATE AND ON BENTHIC COMMUNITY RESPONSES				EFFECTS OF TIME ON MODEL RESIDUALS		
Benthic Community Metric	**Significant variables**	**AIC**	**P**	**Adjusted R^2**	**P**	**Adjusted R^2**	**Slope**
Total Abundance	Mean Chl-a-3mo Lease Gravel	629	**< 0.001**	0.83	0.97	0	0.03
Polychaetes	Gravel Silt	541	**< 0.001**	0.31	0.13	0.02	-0.728
Nematoda spp.	Lease Silt	512	**< 0.001**	0.78	0.65	0	-0.190
Amphipods	Gravel Coarse Sand	516	**0.010**	0.35	**<0.001**	0.16	-1.21
Species Richness	Mean Phosphate-3mo Gravel Coarse Sand	367	**< 0.001**	0.31	**<0.001**	0.15	-0.36
Heteropodarke heteromorpha	Lease	311	**< 0.001**	0.70	0.50	0	-0.05
Hemipodia simplex	Mean Position of X2 TOC	291	**< 0.001**	0.51	0.65	0	0.030
Opheliidae	Gravel Coarse Sand Mean Position of X2	279	**< 0.001**	0.48	0.87	0	-0.010

Figures

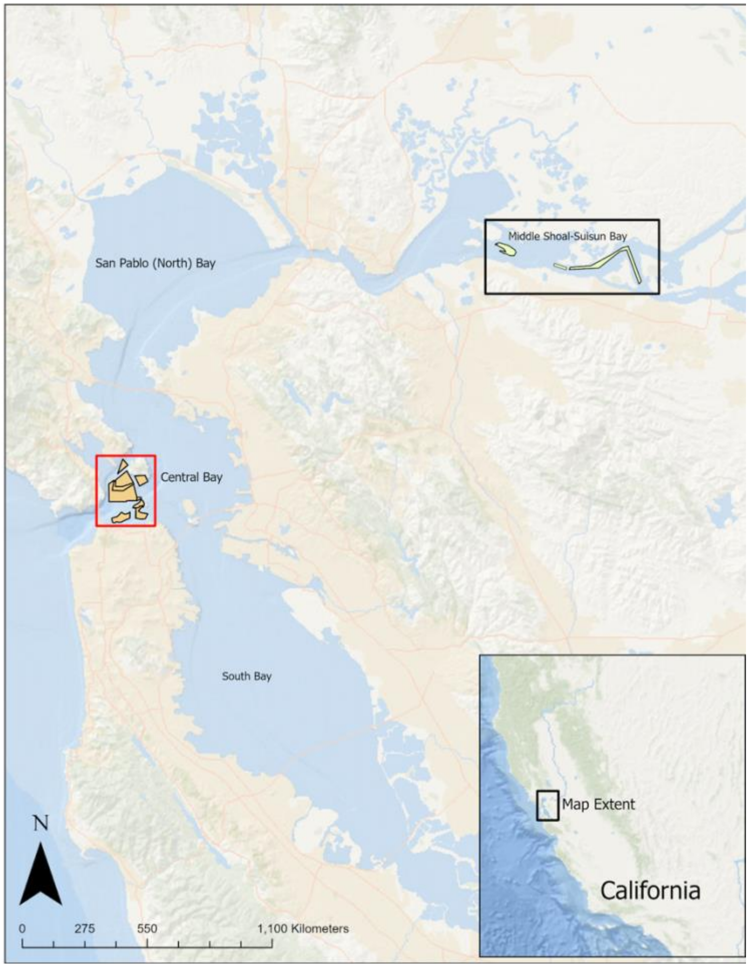


Figure 1. *The four sub-embayments of the San Francisco Bay-Delta region. The red box denotes the area of the Central Bay Sand Mining lease areas (shaded orange). The black box denotes the area of the Middle Shoal-Suisun Bay Sand Mining lease areas (in green). The focus of this paper is the Central Bay lease areas.*

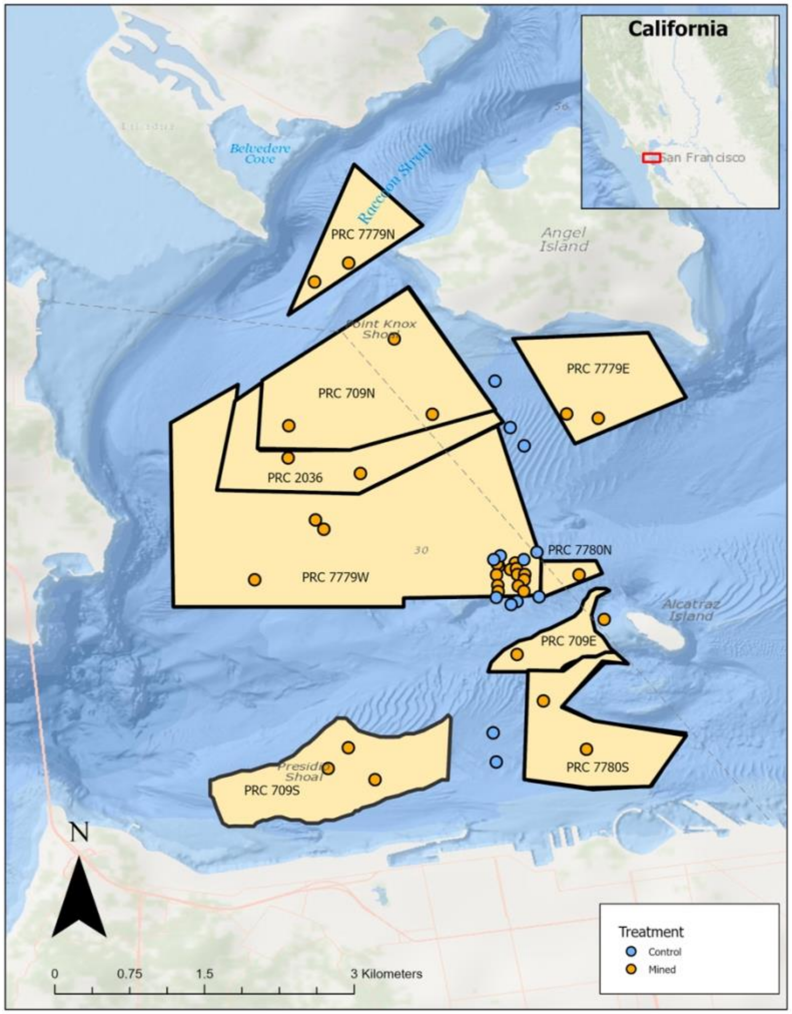


Figure 2. *Sand mining lease areas from Central Bay with benthic survey sampling sites. Orange sites are cited as 'Mined', whereas blue sites are listed as 'Control'.*

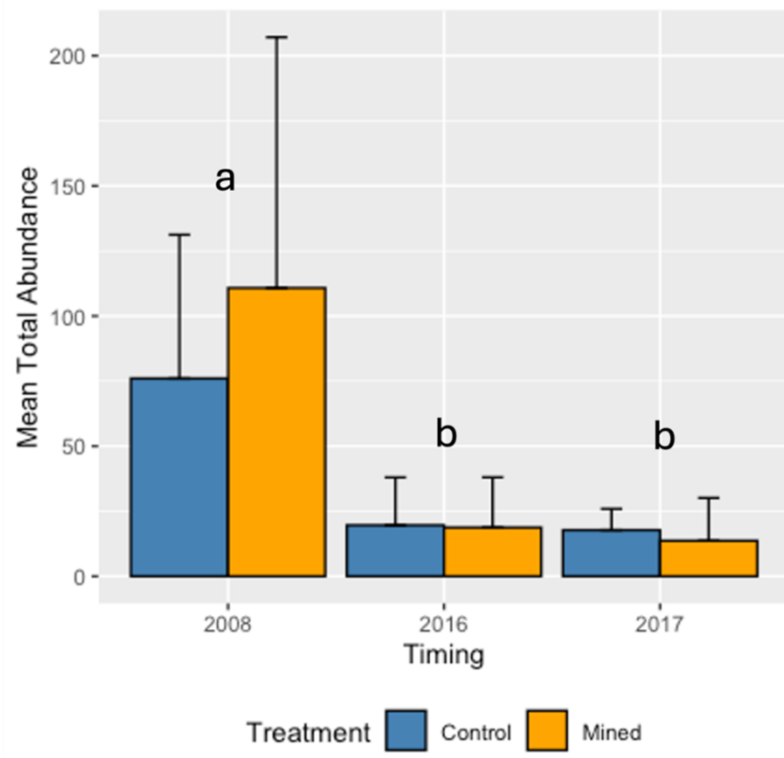


Figure 3. Mean total abundance for all species for all years between control and mined leases. 2008 was significantly different from 2016 (p = <0.0001) and 2017(p = <0.0001). Mean total abundance was not different between 2016 and 2017 (p = 0.96). There was no effect of treatment (p = 0.86).

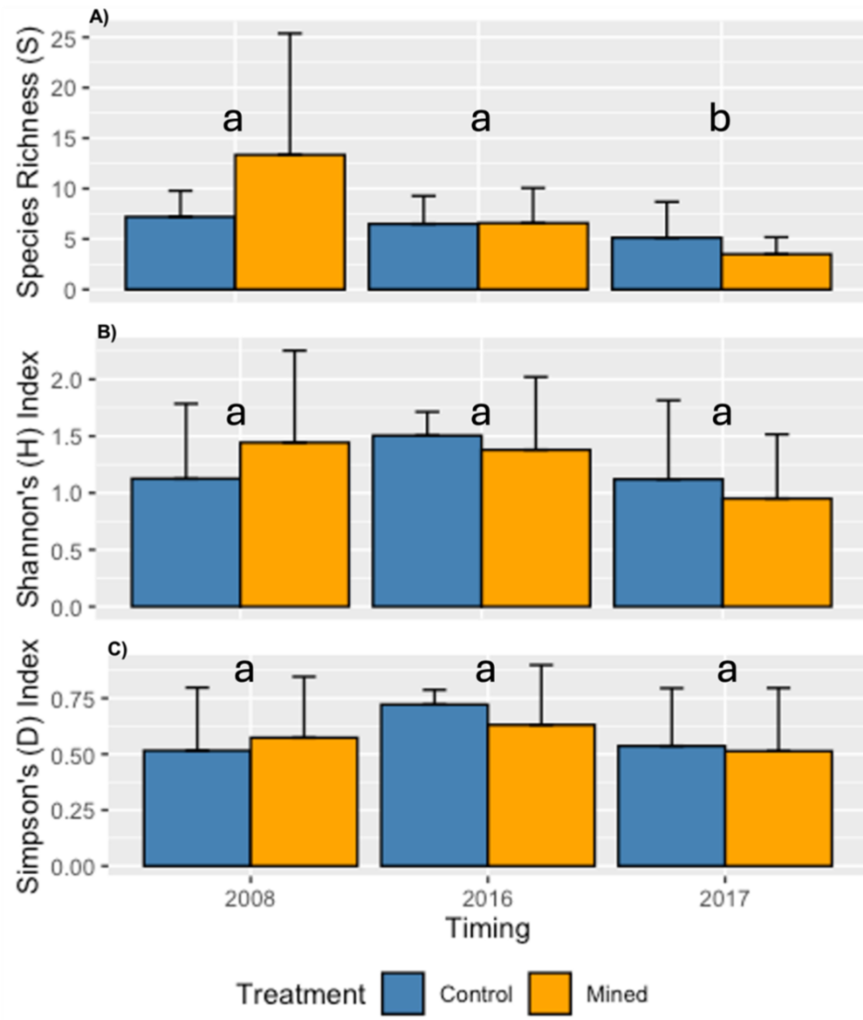


Figure 4. *Mean species diversity metrics for all years between control and mined leases. A) Species Richness, B) Shannon's (H) Index, and C) Simpson's (D) Index. Species richness was significantly different between 2008 and 2017 (p = <0.0001) and between 2016 and 2017 (p = 0.011), there was no difference between treatment (p = 0.427). Neither timing nor treatment were different for Shannon's (H) Index (p = 0.105 and p = 0.897, respectively), and Simpson's (D) Index (p = 0.127 and p = 0.983).*

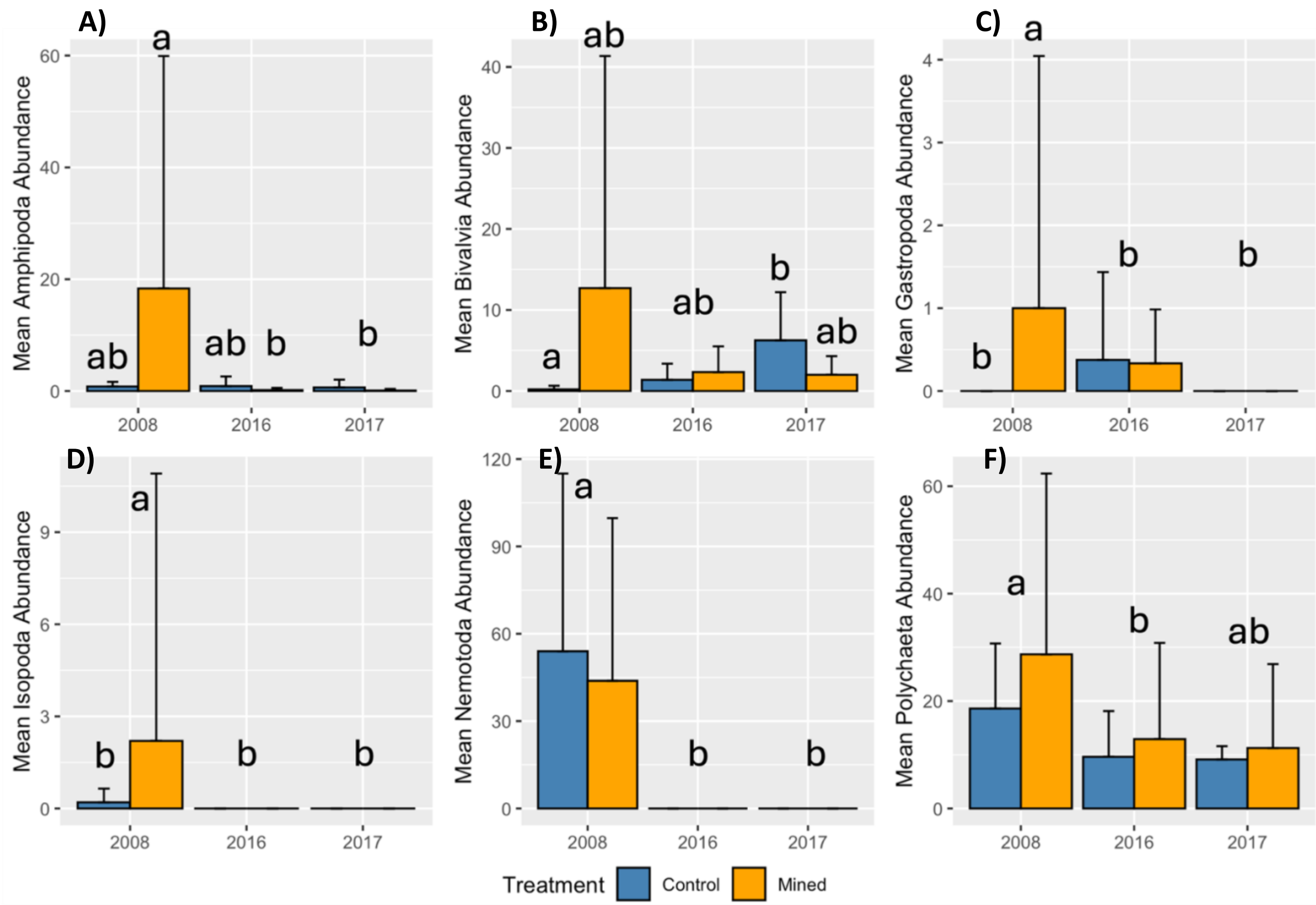


Figure 5. *Mean abundance of select taxonomic groups across years between control and mined leases: A) Amphipoda, B) Bivalvia, C) Gastropoda, D) Isopoda, E) Nematoda, and F) Polychaeta. Amphipod abundance was different between mined areas in 2008 and 2016 (p = 0.0006), and for 2008 mined compared to both 2017 control (p = 0.02) and 2017 mined (p = 0.0001). Bivalve abundance was different between control areas in 2008 and 2017 (p = 0.04). Gastropoda abundance was different for mined areas in 2008 (p = 0.0055). Isopoda abudance was different between treatments in 2008 (p = 0.0001), but not 2016 and 2017. Nemtoda abundance only occured in 2008 which was significantly different than 2016 and 2017 for both mined and unmined areas (timing: p = <0.0001; treatment: p = 0.004). Polychaeta abundance was not different between treatments (p = 0.69) but was different between years where 2008 was different from 2016 (p = 0.037).*

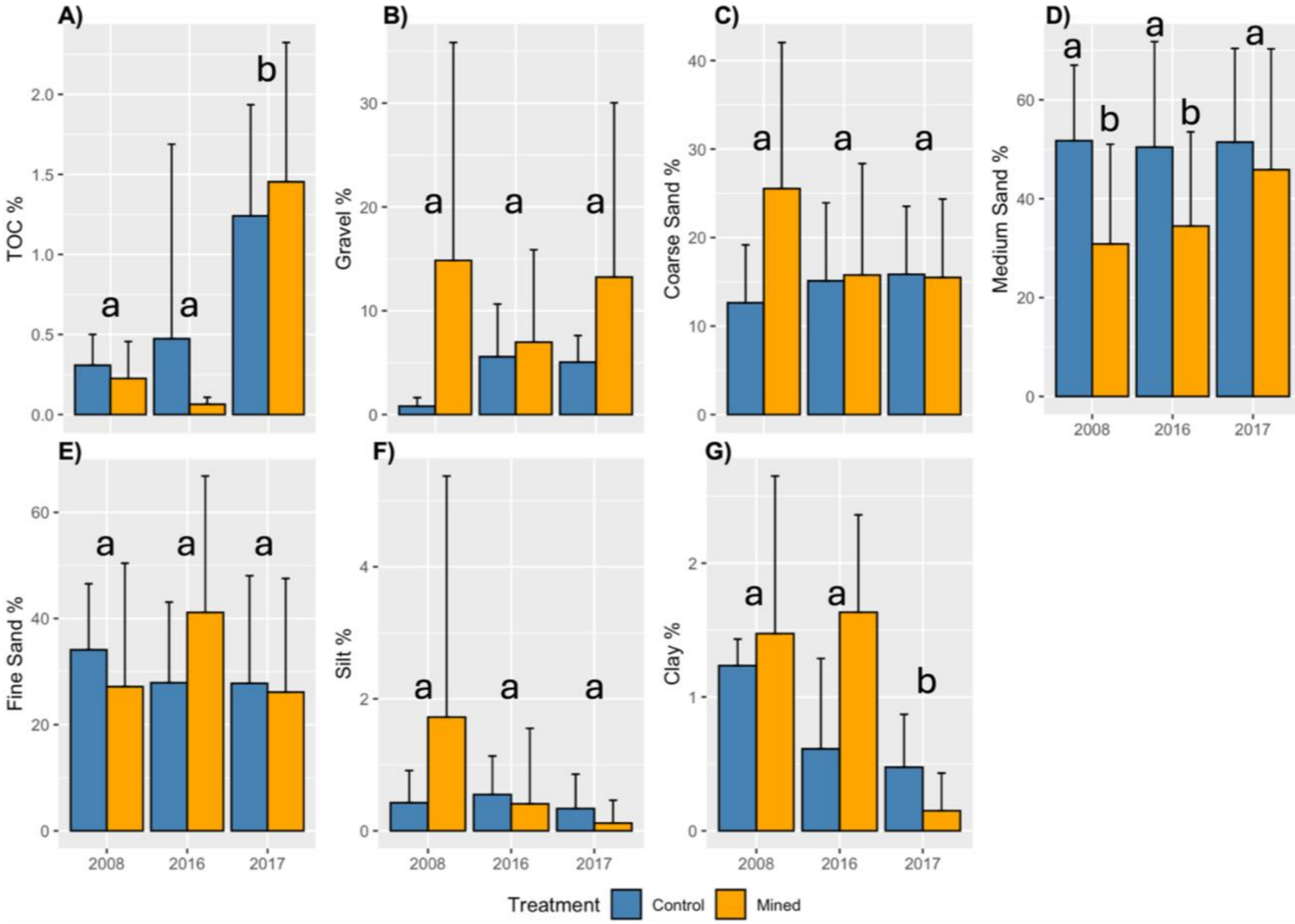


Figure 6. *Comparison of sediment parameters in Central Bay among surveys between control and mined leases. 2017 had significantly different TOC% values compared to 2008 and 2016 (p <0.0001). There was no significant difference between timing and treatment for Gravel% (p = 0.55 and p = 0.128). Coarse sand% was not affected by timing nor treatment (p = 0.376 and p = 0.203), neither was fine sand% (p = 0.256 and p = 0.596) nor Silt% (p = 0.09 and p = 0.298). Medium sand% was significantly different between treatments for 2008 and 2016 (p = 0.007). Clay% was different between years, where 2017 was different from both 2008 and 2016 (p = <0.0001) as well as between treatments (p = 0.047).*

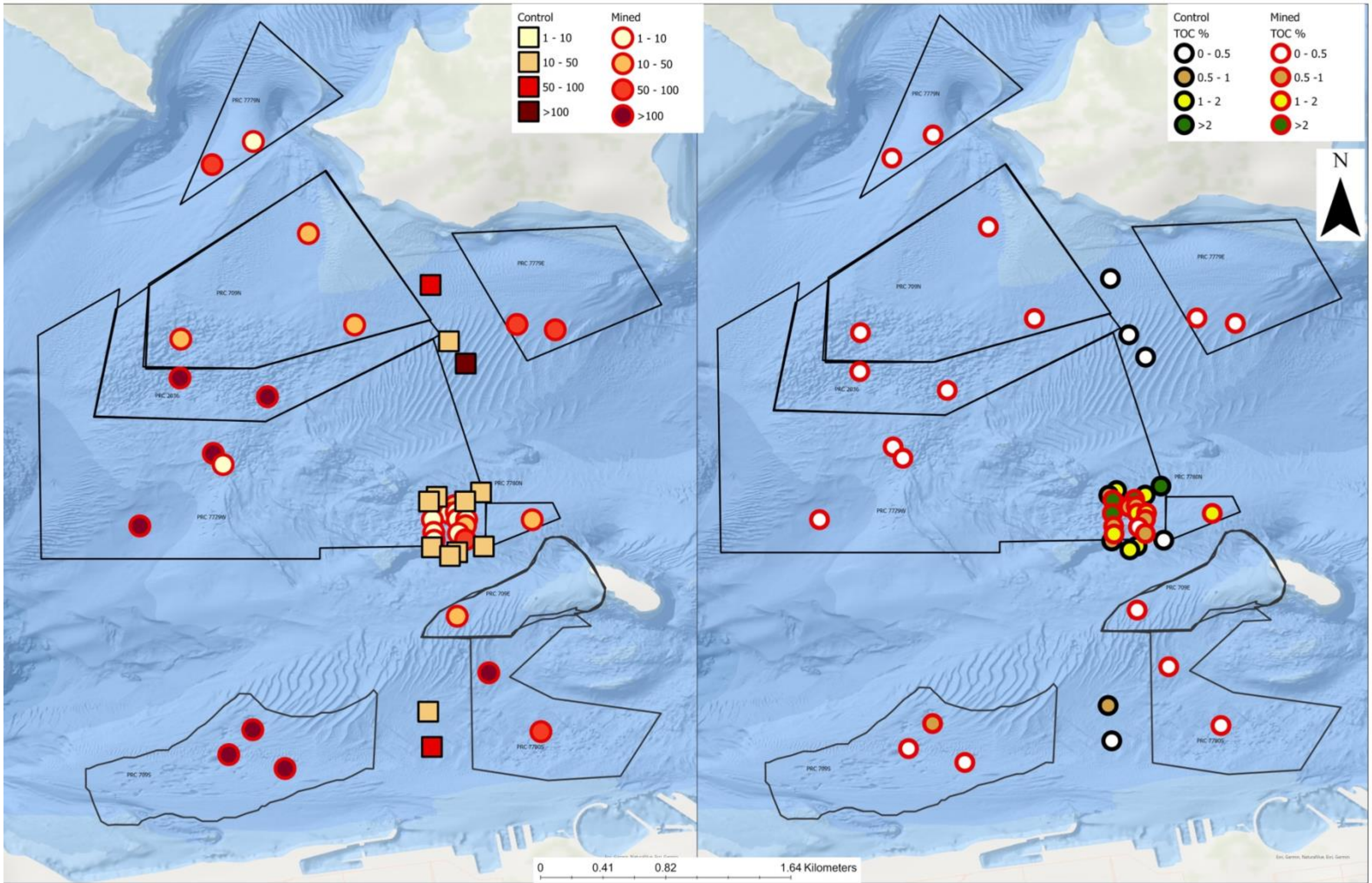


Figure 7. *Spatial patterns in total abundance (left) and TOC content (right) of benthic samples between mined and control areas in Central Bay.*

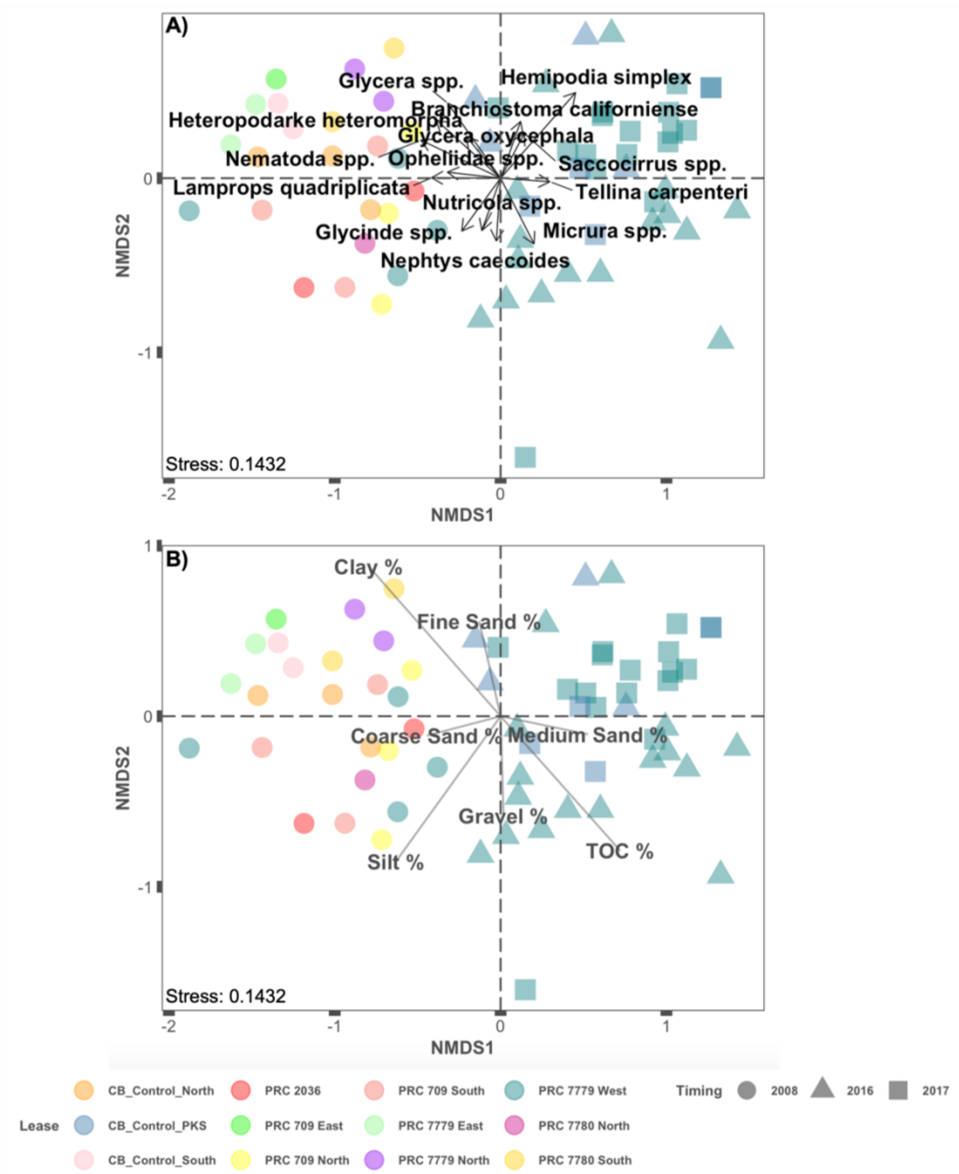


Figure 8. *Results on non-metric multidimensional scaling (NMDS, stress = 0.1432) analysis of benthic communities across mined and control lease sites within Central Bay in 2008 (circle), 2016 (triangle), and 2017 (square). Species ordination (panel A) is over laid onto the plot ordination with arrows indicating the direction of influence of significant species (p-value <0.05). Sediment parameters ordination (panel B) is overlaid onto the plot ordination with arrows indicating the direction of influence for all sediment characteristics.*

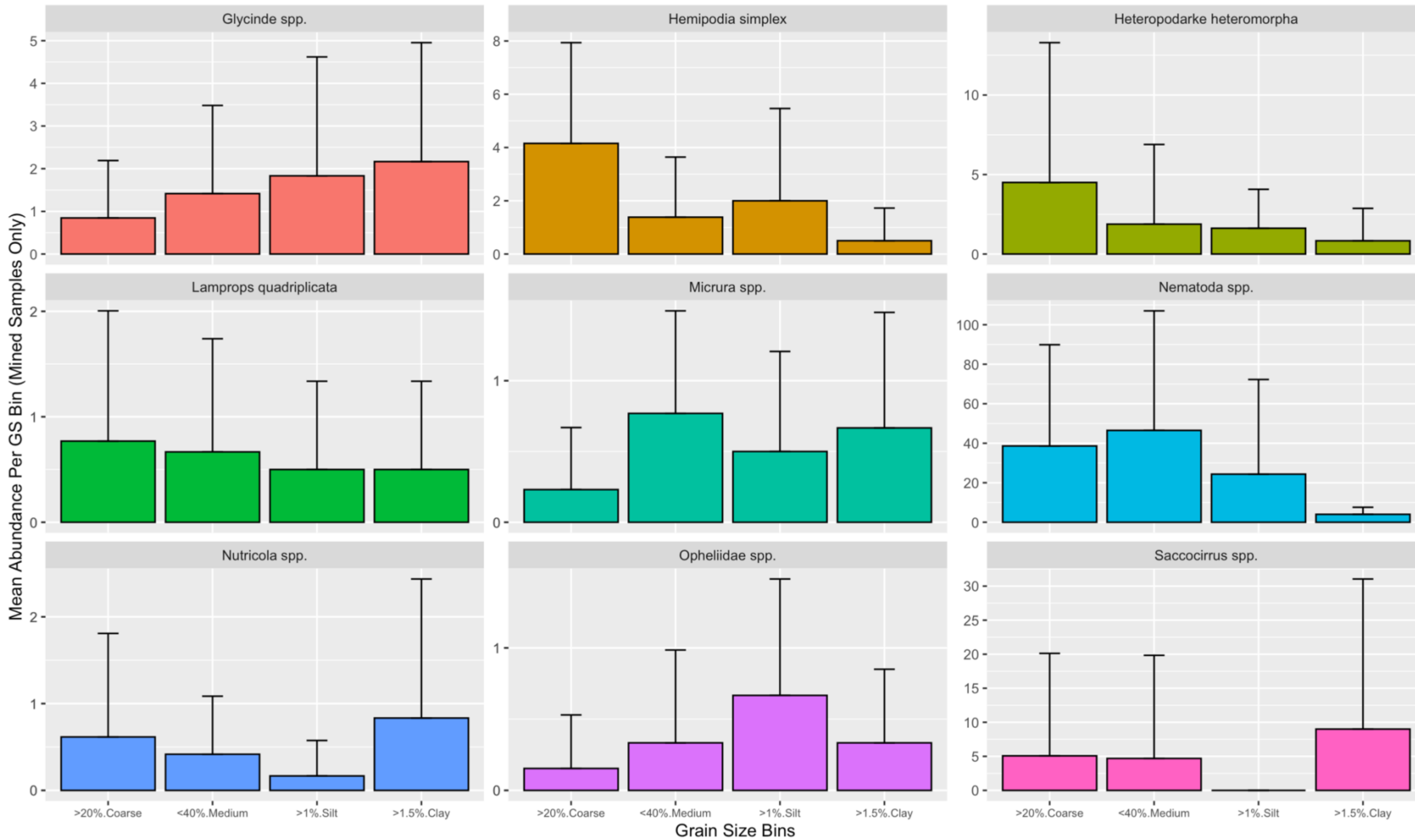
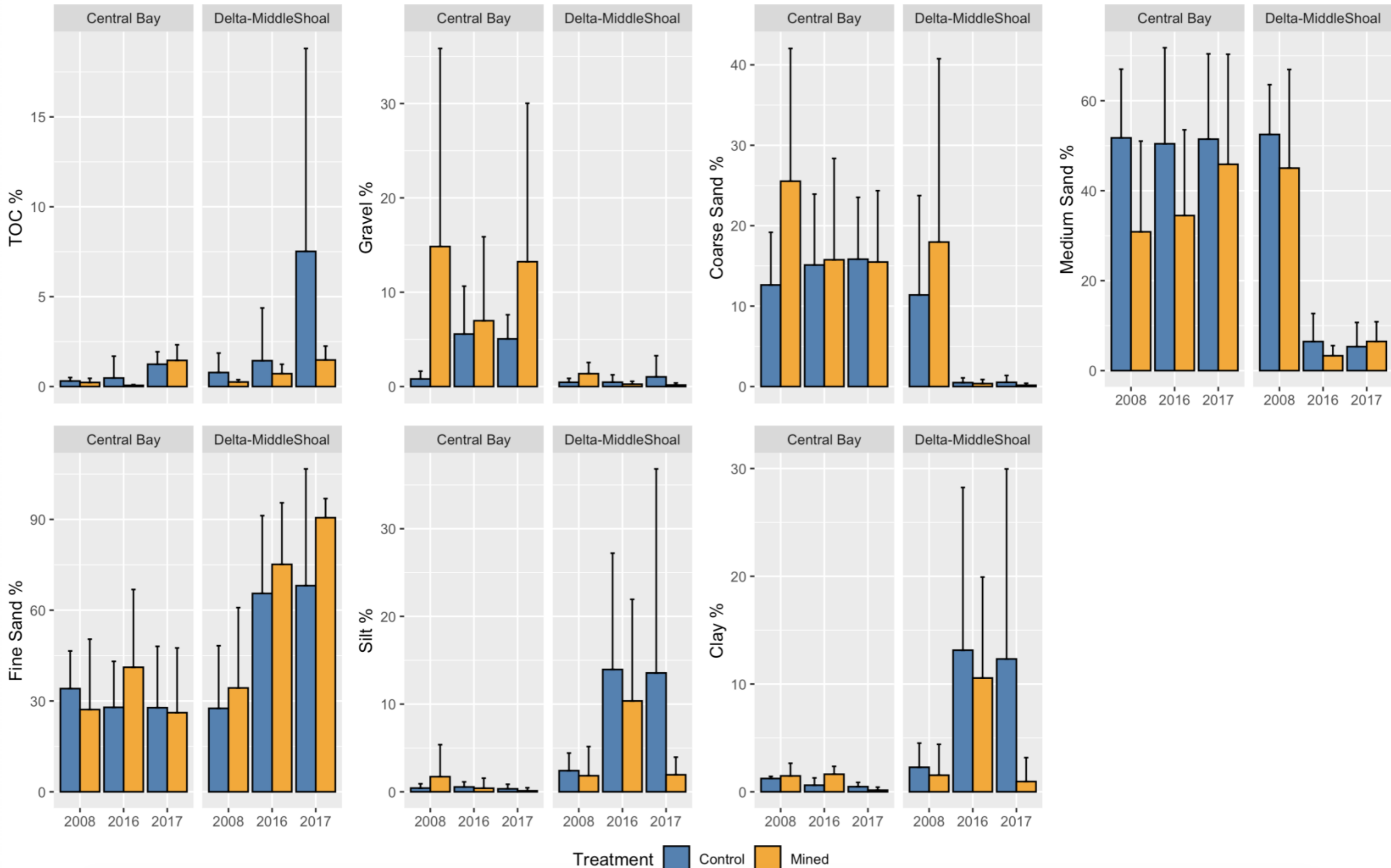


Figure 9. *Mean Abundance of select taxonomic groups in mined samples across years binned into four grain size categories from coarse (>20% Coarse Sand) to very fine (>1.5% Clay) sediments.*

Appendix



Appendix A1. *Comparison of sediment parameters in Central Bay vs Suisun Bay Middle Shoal among surveys between control and mined leases. Due to the vastly different grain size among the surveys of the Suisun Bay Middle Shoal those data were not used in this paper.*