Spatiotemporal patterns and environmental drivers of macrofauna and sediment community oxygen consumption of the Gaoping Continental Shelf off southern Taiwan

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Abstract

Keywords: Authors must provide 4 to 6 keywords plus regional index terms. At least four of the subject keywords should be selected from the Aquatic Science & Fisheries Thesaurus.

1. Introduction

* Background
  + River-influenced oceanic margins (RiOMar)
  + Stats (see thesis)
  + Role of sediment metazoans
* Theories and previous studies
  + Rhoads (1985) proposed that the benthic communities in the River-influenced oceanic margins are controlled by sedimentation stress and nutrient supplementation from the adjacent river. (elaborate)
  + Studies after Rhoads generally echoed Rhoad’s (1985) finding.
  + Loss of diversity and abundance after the river flood.
  + Changes in vertical distribution.
* Current knowledge gaps
  + To our knowledge, little attention was paid to the benthic communities at the oceanic margins adjacent to small mountainous rivers (SMR) (but see Akoumianaki et al., 2013, 2006; Akoumianaki and Nicolaidou, 2007).
  + Due to their small watersheds and steep topographies, SMRs stochastically export mass sediments into the ocean, plays and important role in global carbon cycling.
  + Furthermore, few studies have been conducted to link sediment ecosystem functioning and the marine benthos despite acknowledging their importance in sediment ecosystem functioning.
  + Climate change alters intensity and frequency of extreme weather events.
  + By expanding geographical coverage, increasing temporal resolution, and integrating ecosystem functioning measurements, researchers could better project ecological changes and the associated ecosystem response in future climate scenarios.
* Study area (extract thesis text)
  + Geographical location and geological origin of the Gaoping Continental Shelf.
  + Basic information of the Gaoping River.
  + Climate conditions, dry and wet seasons (monsoon).
  + Sediment discharge patterns of the Gaoping River (annual load).
  + Roles of extreme events, namely typhoons and earthquakes.
  + Source and fate of organic matter in this system.
* Study objective
  + The objective of this study is to document the spatiotemporal variations of the macrofauna standing stock, community assemblage, and the associated sediment oxygen dynamics.
  + Specifically, we are interested in the spatiotemporal changes of macrofaunal abundance, biomass, composition, and the sediment community oxygen consumption (SCOC).
  + We further identified environmental drivers that shaped those features of the soft sediment ecosystem.

1. Material and methods
   1. Shipboard sampling

Two cruises were conducted to collect biological and geochemical data in the Gaoping Continental Shelf (GS) (figure 1). The cruises OR1-1219 and OR1-1242 were conducted in March and October 2019, respectively. S4 was only visited in OR1-1219, while S1 and S2 were visited in OR1-1242. The rest of the four stations, namely S3, S5, S6, and S7, were revisited during the two cruises.

Between the two cruises, one typhoon, namely Typhoon Bailu made landfall in Taiwan on 1 p.m. at August 24th and caused extreme precipitation in southeast and southwest Taiwan (Lin et al., 2020). The extreme precipitation Bailu had brought caused floods and landslides across counties in southern Taiwan. Bailu also damaged harbors, caused death and injuries, and caused agricultural production loss around 170 million NTD (Lin et al., 2020). Despite the immense damage Bailu had caused, the typhoon provided a rare opportunity to examine the response of the benthic communities to extreme weather events.

In each station, Conductivity-Temperature-Depth (CTD) sensors were deployed to collect the profiles of salinity, temperature, water transmission, fluorescence, and dissolved oxygen from the surface water to 5 m above the seafloor. The rosette bottles on the CTD also collected bottom water for sediment incubation. Multi-corers were deployed to collected. Two core tubes were sectioned into 1-cm slices from top to bottom. Each sediment slice was then portioned for analyzing geochemical signatures, such as sediment grain size, sediment porosity, and other bulk sediment compounds. Another three core tubes were chosen by their sediment length and better integrity for shipboard incubation (Glud, 2008).

The three sediment cores were incubated in a temperature-controlled water bath under dark condition to measure the total oxygen utilization (TOU) of the sediment. The water bath temperature was set to be within the range of the bottom water temperatures measured with CTD. Each sediment core was sealed by a plastic lid with a magnetic stir bar attached inside. After removing the air bubbles in the overlying water, another magnetic stir bar attached to a motor was placed above the plastic lid, coupling to the stir bar underneath. Both motor and magnet bars rotated and stirred the overlying water at 60 rpm to prevent water stratification. Four every 4 to 6 hours, we used a miniature oxygen optode (PreSens PSt7) to measure oxygen concentration through a small resealable sampling port and a temperature sensor (PreSens Pt100) for the water bath temperature. The oxygen optode was calibrated for each cruise by scanning the product-specific barcode. We used PreSens Microx 4 data logger to record the dissolved oxygen and temperature until the oxygen levels dropped below 85% of the initial concentration (Glud, 2008). We then used the following equation to estimate TOU (Song et al., 2016): TOU = m \* V/A \* 24/1000, where m is the regression slope, V is water volume, A is area, 24/1000 adjusts the unit to mmol m-2 d-1.

After incubation, a pair of Unisense oxygen microelectrodes and temperature sensors were calibrated by endpoint methods every 24 hours (Unisense A/S, 2020) and lowered into the sediment by a step motor to measure oxygen concentration profiles. The oxygen saturation endpoint was made by air-pumping the seawater with known salinity and temperature for at least 10 minutes. The oxygen depletion endpoint was made by adding sodium dithionite (Na2S2O4), a strong reducing agent, into the seawater with known volume, salinity, and temperature. Sediment oxygen concentration profiling started from roughly 1 cm above the sediment surface, with the step motor moving downward for 100 um per step until the oxygen readings of both the microelectrodes reached zero. At each step, the microelectrodes stopped for 3 seconds to stabilized themselves before taking measurements. According to Fick’s Law of Diffusion, we used the oxygen concentration profiles compensated by temperature, salinity, and sediment porosity to estimate dissolved oxygen utilization (DOU). The sediment oxygen concentration profiles were converted to the diffusive oxygen utilization (DOU) using Unisense Sensor Trace Suite software (Berg et al., 1998). The software calculates the oxygen consumption of the depth profile by intervals and then taken as the best estimator for each core. Another indicator for fauna activities, the oxygen penetration depth (OPD), was defined as the distance from the sediment-water interface to where the oxygen is depleted.

Since both TOU and DOU were measured on board, they were adjusted to *in situ* values using the temperature coefficient Q10 (Valiela, 2015).

Q10 was assumed to be 2, as for most ectotherms; Rincub. and Tincub. were the measured oxygen utilization and incubated temperature; and Tin situ was bottom water temperature. We estimated the contribution of metazoans on TOU, namely the benthic-mediated oxygen utilization (BOU), by subtracting TOU with DOU (Glud, 2008). Since BOU strongly covaried with TOU (r = ), BOU was not included for later analysis. DOU measurements, on the other hand, were generally similar between samples in compare to other regions of the world, and was therefore not included for later analyses (see discussion).

After sediment profiling was completed, the upper 10 cm of the sediment were sieved through a 300 um and washed with filtered seawater to retain macrofauna. The retained samples were then fixed with a 1:1 ratio of filtered seawater and a Rose Bengal-stained 10% formaldehyde solution. The fixed samples were allowed to sit at least a week before sorting and measuring for body size.

* 1. Laboratory analysis

Median grain size (D50), sand fraction, silt fraction, and clay fractions were measured with a laser diffraction size analyzer (Model LS-13320; Beckman Coulter Inc.). Sediment porosity (Por) and water content were measured by calculating the volume and weight difference after a 100℃ for 24 hours assuming a specific density of 1.024 g cm-3 (Blum, 1997).

We measured total organic carbon (TOC), total nitrogen (TN), carbon-to-nitrogen ratio (CN), sediment chlorophyll a (Chla), and stable isotopic carbon (δ13C) of the sediments for bulk material. In brief, the sediment subsamples were first acidified with 6N HCL and then sent to the Flash 2000 elemental analyzer in tandem with a GC-Isolink interface to a Delta V Plus isotope ratio mass spectrometer (both from Thermo Fisher Scientific) to measure TOC and δ13C. TOC values were reported in percent weight contribution, while δ13C values were calculated using Vienna-PeeDee Belemnite as the reference point and reported in ‰ differences. TN measurements were similar to TOC and δ13C except no acid pretreatments were applied before sending the sediment samples into the same instruments. CN was then derived by dividing TOC with TN. For Chla, the sediment samples were first sitting in 10-ml acetone-water solution with a volumetric ratio of 9:1 in the dark at 4℃ for 24 hours. The chlorophyll extracts were then measured with a fluorometer (Model 10-AU; Turner Designs, Inc.) and reported in nanogram per gram dry weight.

Back in the lab, the liquid of the fixed fauna samples was replaced with 70% ethanol for long-term storage. I sorted the fauna samples into major taxa (mostly at order level) and measured their relevant body dimensions with an ocular micrometer for biomass estimation. While specimens with at least on of its main body parts (i.e., head, thorax, and abdomen) were measured, only the specimens with their cephalic region intact were included in later analysis.

Various methods were used to estimate taxon-specific biovolume. The biovolumes of polychaetes were estimated with each individual’s body shape. The body shapes of vermiform polychaetes were assumed to be cylinder, while sternaspid polychaetes were assumed to be ellipsoids. The biovolumes for some common benthic taxa were calculated using the Length-Width Relationship (LWR): V =cLW2, where V is the individual volume; c is the taxon-specific conversion factor; L and W are the maximum length and width of the individual (Feller and Warwick, 1988). The biovolumes of some odd-shaped taxa were also estimated using LWR with conversion factors borrowed from taxa with similar body shapes. The biovolumes of some other taxa with relatively simple body shapes were approximated with geometric shapes (i.e., cylinder, cone, ellipsoid) (e.g., Hillebrand et al. 1999). The rest of the taxa were assumed to be cylindrical during measurement following the general volumetric method (Benoist et al., 2019). The derived biovolumes were then converted to wet weight by assuming a specific density of 1.13 (Gerlach et al., 1985).

* 1. Data analysis

All data analysis were conducted using R version 4.0.3 (R Core Team, 2020) with additional statistical packages *vegan* (Oksanen et al., 2021), *MuMIn* (Bartoń, 2022). Packages *dplyr* and *tidyr* were used for data cleaning (Wickham, 2021; Wickham et al., 2021). Results were visualized with *ggplot2* and its extensions (Schloerke et al., 2021; Slowikowski, 2021; Wickham, 2016).

We divided explanatory variables into (1) spatiotemporal factors and (2) environmental variables to analyze spatiotemporal patterns and identify environmental drivers separately. Despite not directly measured, the DRM of each station was calculated using World Geodetic System 1984. The coordinate of the river mouth was arbitrarily set at 120.423960⁰E and 22.470504⁰N using Google Map (figure 1). Key environmental variables were screened out with ecological reasoning and a Pearson correlation coefficient threshold of |r| > 0.7 to prevent spurious results (Dormann et al., 2013). Dissolved oxygen was excluded since all stations were well-oxygenated (Middelburg and Levin, 2009). Strong correlations were present between salinity, transmission, and temperature (|r| > 0.7); temperature was selected into further analysis due to its cross-scale ecological importance (Brown et al., 2004). Due to strong collinearity between clay, silt, sand fractions, and D50 (|r| > 0.7), D50 was selected as the sole indicator of sediment granulometry. Porosity and TN were highly correlated (r > 0.7). Since other geochemical indicators, such as CN and Chla, also indicate organic matter quality (Campanyà-Llovet et al., 2017), TN was excluded in later analyses. δ13C and Chla were negatively correlated (r = -0.76). Chla was included for further analyses since δ13C signatures may be confounded by marine autochthonous carbon, modern terrestrial carbon, and petrogenic carbon in the present study area (Carter et al., 2012; Kao et al., 2006). The resulting variables were *Temp*, *Fluo*, *Por*, *D50*, *TOC*, *CN*, and *Chla*. The variables were centered and scaled to unit variance. Two axes of the principal component analysis (PCA) were extracted to visualize the environmental difference among sampling sites.

Macrofauna abundance and biomass assemblages were Box-Cox-chord transformed (Legendre and Borcard, 2018). While Hellinger transformation strictly takes the exponent of 0.5 on ecological data before chord transformation, Box-Cox-chord transformation is more flexible, providing a series of exponents for ecologists to choose from. Using the Dagnelie test, the exponent that reaches multinormality of the Mahalanobis generalized distance can be sought (Legendre and Borcard, 2018). In this study, the exponent for macrofauna abundance and biomass compositions were 0.3 and 0.1, respectively. Euclidean distance was then used to calculate between-sample pairwise distances. The first two axes of the principal component analysis (PCA) were used to visualize the compositional difference between samples. Only taxa with 40% of their variation explained by the first two PCs were shown. On the other hand, macrofauna abundance and biomass were log10 transformed, while TOU were not., while scatterplots were drawn to show spatiotemporal variations between stations.

Water depth, the distance between each station and the Gaoping River mouth (DRM), and Cruise with all possible two-way interactions were fitted on (1) macrofauna abundance composition, (2) biomass composition, (3) abundance, (4) biomass, and (5) TOU to examine spatiotemporal patterns. We used PERMANOVA with 9999 permutations on macrofauna abundance and biomass compositions (Anderson, 2017), while we fit simple linear regressions on macrofauna abundance, biomass, and sediment TOU under the multimodel inference framework. Preventing arbitrarily lean on a single model, the multimodel inference framework uses information theoretic approaches to account uncertainty across several models, identifies competing models, and then draws inferences from them (Burnham and Anderson, 2002). We sought all possible combinations of the explanatory variables and extracted an un-nested top model set with ΔAICc < 6 (Richards, 2008, 2005). Using such a conservative criteria has the advantage of having few parsimonious models for inference (Richards, 2008). We then took the natural average of the resulting models to seek possible ecological trends (Burnham and Anderson, 2002).

Environmental variables were fitted on macrofauna abundance composition, biomass composition, abundance, biomass, and TOU to identify potential drivers, except that we assume no interactions between those variables. Redundancy analysis (RDA) with p-value-based backward selection were used to retain a set of environmental variables that best described the compositional data. Similar to what we described above, macrofauna abundance, biomass, and TOU were also fitted with environmental variables with simple linear regression under a multimodel framework.

1. Results
   1. Environmental condition and macrofauna

PC1 and PC1 of the environmental variables explained 36.6 and 25.6% of the total variation (figure 2). PC1 was loaded with high contributions of Temp, Fluo, Chla, and CN. PC2 was loaded with TOC, Por, and D50. Shallower stations of OR1-1242 (S2, S3, S6, and S7) were aggregated at the right-hand side of the PCA figure, while deeper stations in OR1-1242 and all stations in OR1-1219 aggregated at the left-hand side of the PCA.

* 1. Spatiotemporal patterns and environmental drivers of macrofauna composition

Polychaetes dominates the abundance (figure 3).

PERMANOVA results on macrofauna abundance composition (Table ).

PC1 and PC2 of the macrofauna abundance composition explained 18.14 and 15.94% of the total variation (figure 4a). More the 40% of the variation of peracarids (isopods, tanaids, and amphipods), aplacophorans, and hydroids were explained in the first two PCs. The distribution of the samples were less clear

PC1 and PC2 of the macrofauna biomass composition explained 20.57 and 13.69% of the total variation (figure 4b). More the 40% of the variation of aplacophorans, mysids, ophiuroids, and decapods were explained by the first two PCs. The distribution of samples greatly corresponds with the taxa vectors. The samples of most sites in OR1-1219 and deeper sites in OR1-1242 (S1, S5) are characterized with high contributions of ophiuriods and aplacophorans. Shallower sites in OR1-1242 (S2, S3, S6, and S7) are characterized with high contributions of mysids and decapods.

RDA explained 31.44% of macrofauna abundance composition’s variation, while RDA1 and RDA2 explained 40.85 and 27.65% of that variance, respectively (figure 5a). *D50*, *Por*, *Chla*, *TOC*, and *CN* were retained in the final RDA model. More than 40% of the variation of oligochaetes, mysids, and aplacophorans were explained by the first two RDA axes.

RDA explained 34.35% of macrofauna biomass composition’s variation, while RDA1 and RDA2 explained 43.02 and 26.04% of that variance, respectively (figure 5b). *D50*, *Temp, Por*, *Chla*, *TOC*, and *CN* were retained in the final RDA model. *Temp*, *TOC*, *Chla*, and *CN* contributed greatly in RDA1, while D50 and Por contributed greatly in RDA2. More than 40% of the variation of Decapods, mysids, and aplacophorans were explained by the first two RDA axes.

* 1. Spatiotemporal patterns and environmental drivers of macrofauna abundance, biomass, and TOU

The model average results of spatiotemporal variables generally echoed with the scatter plot (figure 6).

* Number of models in the top model set
* Significant/ non significant variables

1. Discussion
   1. Study limitation

The estimated DOU in the present study might not be representative to in situ DOU. DOU measurements in the present study were low in compare to other studies at similar habitats. *In situ* DOU measurements off the Rhone river have shown that oxygen profiles and the estimated DOU can have huge variations within days after sediment resuspension events (Toussaint et al., 2014). The Gaoping Continental Shelf seafloor receives inputs from the Gaoping River and is subjected to tidal waves (Liu et al., 2002). Transmission profiles in OR1-1242 showed peaks near the seafloor, indicating higher concentrations of suspended particles in the bottom water (CTD figure). Moreover, oxygen profile measurements in the present study were generally done more than 24 hours after retrieval, meaning that the measured DOU might not reveal in situ conditions. We therefore suggested that the DOU in the present study is systematically lower than the in situ DOU at core retrevial. We warn that incubation experiments might stabilize the sediment, lowering the measured DOU, yielding erroneously high BOUs as a result. Also, future studies should specify when their oxygen profiles were measured.

* 1. Environmental patterns in the Gaoping Continental Shelf

The distribution of the samples in both RDAs showed similar patterns, suggesting that environmental changes alters in macrofauna abundance and biomass compositions.

1. Conclusions
2. Acknowledgements
3. References

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