



# Biogeochemical responses to ocean alkalinity enhancement in the North China Sea

Xiaoke Xin<sup>1</sup>, Yimeng Liu<sup>1</sup>, Ruize Chang<sup>1</sup>, Ruobin Lei<sup>1</sup>, Minghao Zhu<sup>1</sup>, Xiaotong Wang<sup>1</sup>, Xue Chao Wang<sup>1</sup>, Jihua Liu<sup>1,2</sup>

5 <sup>1</sup>Institute of Marine Science and Technology, Shandong University, Qingdao 266237, China.

<sup>2</sup>Global Ocean Negative Carbon Emissions (ONCE) Program Alliance, Qingdao, China.

*Correspondence to:* Jihua Liu (liujihua1982@foxmail.com)

**Abstract.** Ocean Alkalinity Enhancement (OAE) has been proposed as a promising marine carbon dioxide removal strategy. Yet its ecological consequences remain poorly understood, particularly in dynamic coastal systems characterised by high environmental variability. Here, we investigated the effects of OAE on plankton community structure and biogeochemical processes in the eutrophic North China Sea (NCS) using a microcosm experiment with natural seawater collected offshore. Two OAE scenarios were evaluated: CO<sub>2</sub>-equilibrated and CO<sub>2</sub>-non-equilibrated treatments, both targeting an increase in total alkalinity ( $\Delta$ TA) of approximately 500  $\mu\text{mol kg}^{-1}$ . Neither OAE treatment significantly affected the abundance of phytoplankton and microzooplankton. Similarly, plankton communities exposed to OAE maintained their natural levels of chlorophyll a, taxonomic composition, biodiversity, and particulate organic carbon throughout the experiment. In contrast, substantial TA loss occurred in the CO<sub>2</sub>-non-equilibrated OAE treatment, indicating reduced alkalinity retention under non-equilibrated conditions. This study was conducted within the framework of the Ocean Alkalinity Enhancement Pelagic Impact Intercomparison Project (OAEPIIP), which was established to standardise OAE microcosm experiments and systematically assess the environmental impacts of OAE across diverse marine ecosystems. Our findings indicate that NCS phytoplankton and microzooplankton exhibited high tolerance to both CO<sub>2</sub>-equilibrated and non-equilibrated OAE scenarios under the experimental conditions tested, while highlighting the need to monitor and mitigate TA loss alkalinity loss during OAE implementation in warm coastal environments.

## 25 1. Introduction

Anthropogenic carbon dioxide (CO<sub>2</sub>) emissions have intensified global warming and ocean acidification, posing severe threats to marine ecosystems and biodiversity (IPCC, 2023). Ocean Alkalinity Enhancement (OAE) has emerged as a promising negative emissions technology (NET) to sequester atmospheric CO<sub>2</sub>, supporting the Paris Agreement goals of limiting warming to well below 2 or 1.5 °C (Köhler et al., 2010; Renforth and Henderson, 2017). Beyond carbon dioxide removal (CDR), OAE holds the potential to regionally mitigate ocean acidification (Schulz et al., 2023). OAE operates by increasing seawater total alkalinity (TA), thereby increasing the ocean's capacity to absorb atmospheric CO<sub>2</sub> and promoting the conversion of dissolved CO<sub>2</sub> into bicarbonate (HCO<sub>3</sub><sup>-</sup>) and carbonate (CO<sub>3</sub><sup>2-</sup>) ions (Bach et al., 2019; Renforth and Henderson, 2017). This process can lower seawater partial pressure of CO<sub>2</sub> (pCO<sub>2</sub>), increasing the air–sea CO<sub>2</sub> gradient and facilitating additional atmospheric CO<sub>2</sub> uptake while reducing CO<sub>2</sub> efflux from the ocean. The absorbed carbon can subsequently be stored in the ocean over geological timescales (Ilyina et al., 2013).

Despite its potential climate benefits, OAE may alter marine plankton communities, which play fundamental roles in marine food webs and biogeochemical cycling (Bach et al., 2019). OAE induces dynamic shifts in the carbonate chemistry in seawater that may impact marine organism physiology, community composition, and trophic interactions during and/or after equilibration with the atmosphere (Oschlies et al., 2023; Renforth and Henderson, 2017). Existing studies report variable biological responses, ranging from substantial shifts in plankton biomass and community structure to negligible ecological effects under different OAE scenarios (Guo et al., 2025; McElhany et al., 2026; Sánchez et al., 2024; Xin et al., 2024). These discrepancies likely arise from differences in environmental conditions, plankton community composition,



and experimental design, complicating environmental risk assessment and the development of responsible large-scale OAE deployment strategies (Eggers et al., 2014; Kousoulas et al., 2025; Subhas et al., 2022). To improve comparability among studies, the Ocean Alkalinity Enhancement Pelagic Impact Intercomparison Project (OAEPIIP) was established to standardise experimental protocols and enable systematic evaluation of plankton responses across diverse marine environments (Bach, 2024).

The North China Sea (NCS) represents an ecologically important yet understudied region for OAE impact assessment, owing to its distinctive hydrology, ecological complexity, and intense anthropogenic pressure (Tak et al., 2022). The region is governed by multiple coastal current systems that drive strong fluctuations in temperature, salinity, nutrient availability, and water column mixing. Intensive aquaculture and fishing activities further contribute to coastal eutrophication and support productive plankton assemblages and complex coastal ecosystems (Wang et al., 2021). Together, these attributes create a dynamic environmental mosaic in which plankton responses to OAE may deviate substantially from those observed in oligotrophic or open-ocean systems, highlighting the need for region-specific empirical data to evaluate the feasibility and ecological safety of OAE.

Here, we investigated the responses and biogeochemical processes of plankton community to CO<sub>2</sub>-equilibrated and non-equilibrated OAE treatments in coastal waters of the NCS. Our study followed OAEPIIP-standardised methods to ensure consistency and comparability within the global intercomparison framework. Specifically, we aimed to (1) evaluate the ecological effects of OAE on coastal plankton communities in the NCS, and (2) contribute region-specific data to the global OAEPIIP assessment of pelagic ecosystem responses to OAE.

## 2. Materials and Methods

### 2.1 Seawater collection

Surface seawater was collected at a depth of 0.5 m during high tide near Aoshan Bay (36.33°N, 120.72°E), in the North China Sea, in summer 2024. At the time of collection, the Subei Coastal Current influenced local hydrographic conditions and contributed to the transport of nutrients and plankton. Natural plankton communities were gently filtered through a 3 mm mesh to remove large debris and nekton, and subsequently enclosed in 55L PET Kegland® Fermzilla conical unitank fermenters, which served as microcosms for the experiment. All microcosms were transported to the laboratory within 2 hours of enclosure to minimise environmental disturbance. Upon arrival, plankton communities were acclimated to laboratory conditions (12:12 light:dark cycle, light intensity of 150 μmol photons m<sup>-2</sup> s<sup>-1</sup>) for 24 hours prior to the initiation of the experiment. Temperature was maintained at the mean in situ temperature over the course of the study in a temperature-controlled room. For a comprehensive description of the experimental design and technical details, please refer to Bach et al. (2024).

### 2.2 Treatment design

Three treatments, each with triplicate microcosms, were established: (1) Control: untreated seawater, no alkalinity addition; (2) CO<sub>2</sub>-non-equilibrated OAE (nEq-OAE): seawater amended with sodium hydroxide (NaOH) to increase TA by 500 μmol kg<sup>-1</sup>, without equilibration with atmospheric CO<sub>2</sub>; and (3) CO<sub>2</sub>-equilibrated OAE (Eq-OAE): seawater amended with sodium bicarbonate (NaHCO<sub>3</sub>) to increase TA by 500 μmol kg<sup>-1</sup>. Convection-driven mixing was induced within the enclosed microcosms to maintain plankton in suspension, using two 30 W heat belts attached at the bottom of each microcosm. Microcosm placement was shuffled daily to ensure similar light intensity across all units. The target TA enhancement was determined based on both the relevance of a simulated OAE perturbation at the timescales and the detectability of biological effects.

### 2.3 Seawater sampling and measurement

Samples were collected from all microcosms between 08:00–10:00. Prior to sampling, all microcosms were gently stirred to ensure homogenisation. The sampling schedule for each parameter is provided in Table 1. The total sample volume was limited to less than approximately one-third of the microcosm over the



course of the study to limit the build-up of a headspace and maintain a consistent heat input per litre of enclosed volume via the heat belts.

95 Samples for carbonate chemistry and macronutrient analyses were filtered through syringe filters (0.45 µm, Millipore) and dispensed into high-borosilicate amber glasses without headspace for pH measurements and into HDPE bottles for TA determination, respectively. TA was measured using an automatic potentiometric titrator following the method described by Chen et al. (2022). The pH samples were measured using a pH meter A211 (Thermo Fisher, USA) equipped with an Orion electrode 8157 BNUMD (Thermo Fisher, USA). pCO<sub>2</sub> was calculated based on TA and pH data using K1 and K2 equilibrium constants (Lueker et al., 2000). Filtered water samples for nutrient analysis were collected and stored in centrifuge tubes at -20 °C until analysis. After thawing to room temperature, samples were measured using a continuous-flow analyser (AA3, SEAL Analysis Ltd., Germany).

100

105 Samples for chlorophyll a (Chla) and biogenic silica (BSi) were collected by filtration under mild vacuum pressure (-200 mbar). Chla was sampled by filtering seawater through glass-fibre filters (GF/F, 25 mm diameter, 0.7 µm pore size), extracting pigments with 90% acetone at 4 °C in the dark for 24 hours, and measuring fluorescence using Cary Eclipse spectrofluorometer (Agilent Technologies, Santa Clara, CA). BSi was collected by filtering onto mixed cellulose ester (MCE) membrane filters (25 mm diameter, 0.8 µm pore size) and stored at -20 °C prior to measurement. Biogenic silica was determined following the method described by Mortlock and Froelich (1989). In brief, an appropriate amount of seawater sample was sequentially treated with 10% H<sub>2</sub>O<sub>2</sub> and 1 mol L<sup>-1</sup> HCl to remove organic matter and carbonates, respectively. The residual material was then subjected to extraction with 2 mol L<sup>-1</sup> Na<sub>2</sub>CO<sub>3</sub> solution at 85°C to solubilise BSi, and the resulting silicate concentration was measured spectrophotometrically.

110

115 Flow cytometry samples for phytoplankton and bacteria were collected from the microcosms with pipettes and fixed with 20 µL of glutaraldehyde. After mixing samples with fixatives, samples were stored at -80 °C until measurement using a flow cytometer (Accuri C6). Plankton were enumerated using a light microscope (Nikon ECLIPSE Ts2) with a sedimentation chamber (Utermöhl, 1958). Well-mixed seawater samples (25 mL) were fixed with Lugol's solution (final concentration, 1%). Phytoplankton and microzooplankton were counted either across the entire chamber or until at least 200 individuals per sample were enumerated to ensure statistical reliability.

120

Particulate organic carbon (POC) and particulate organic nitrogen (PON) were determined by filtering seawater through pre-combusted (450 °C for 4 h) GF/F filters (Whatman, 0.7 µm). Filters were fumed with concentrated hydrochloric acid vapour for 24 hours to remove inorganic carbon and subsequently dried at 60 °C for 48 h, weighed, and analysed using an elemental analyser (ECS4024, NC-Tech).

125

On the initial and final days, 1 L seawater sample from each microcosm was filtered through 0.2 µm pore size polycarbonate membrane filters (47-mm diameter; Millipore) for microbial community analysis. Total DNA was extracted from 0.5 g of sample using the DNeasy PowerSoil Kit (Qiagen, Germany) according to the manufacturer's protocol. DNA quality and concentration were assessed via NanoDrop2000 spectrophotometry, and DNA integrity was verified by 1% agarose gel electrophoresis. The bacterial 16S rRNA gene V4-V5 hypervariable region was amplified using primers 515F (5'-GTGYCAGCMGCCGCGGTAA-3') and 907R (5'-CCGTCAATTCMTTTRAGTTT-3'), while the V9 region of the eukaryotic 18S rRNA gene was targeted using primer 18SV9F (5'-CCCTGCCNTTTGTACACAC-3') paired with its corresponding reverse primer (5'-CCTTCNGCAGGTTACCTAC-3'). Triplicate PCR products per sample were pooled, verified on 2% agarose gels, purified with the AxyPrep DNA Gel Extraction Kit, and quantified using a Quantus<sup>TM</sup> Fluorometer prior to equimolar pooling. Libraries were constructed with the NEXTFLEX Rapid DNA-Seq Kit on DNBSEQ-T7 platform. Raw reads were quality-filtered and taxonomically assigned using QIIME 2 (version 2025.10). 16S rRNA gene sequences were classified using the SILVA database (v138), while 18S rRNA gene sequences were annotated using the PR2 database (v5.11).

130

135

Table 1. Sampling schedule for each parameter



| Parameters        | Day of experiment |   |   |   |   |   |   |   |   |   |    |    |    |    |
|-------------------|-------------------|---|---|---|---|---|---|---|---|---|----|----|----|----|
|                   | 0                 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 |
| TA                | ✓                 |   | ✓ |   | ✓ |   | ✓ |   | ✓ |   | ✓  |    |    | ✓  |
| DIC               | ✓                 |   | ✓ |   | ✓ |   | ✓ |   | ✓ |   | ✓  |    |    | ✓  |
| pH and Temp       | ✓                 | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓  | ✓  | ✓  | ✓  |
| Salinity          | ✓                 |   |   |   |   |   |   |   |   |   |    |    |    | ✓  |
| Light             | ✓                 |   |   |   |   |   |   |   |   |   |    |    |    | ✓  |
| N, P and Si       | ✓                 |   | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓  |    |    | ✓  |
| BSi, POC&PN, Chla | ✓                 |   | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓  |    |    | ✓  |
| FC                | ✓                 |   | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓ | ✓  |    |    | ✓  |
| Microscopy        | ✓                 |   | ✓ |   | ✓ |   | ✓ |   | ✓ |   | ✓  |    |    | ✓  |
| Metagenomics      | ✓                 |   |   |   |   |   |   |   |   |   |    |    |    | ✓  |

2.4 Statistical analysis

All data are presented as mean ± standard deviation (SD) of triplicate samples. Statistical differences between treatments and the control were tested using Generalised Additive Mixed Models (GAMMs) implemented in the package “mgcv”, following the OAEPIIP-standardised R-based pipeline (OAEPIIP, 2024). Non-metric multidimensional scaling (NMDS) ordination was applied to assess bacterial community composition based on 16S rRNA gene amplicon sequences, as well as microphytoplankton and microzooplankton communities based on microscopy data, using the “vegan” package. All statistical analyses were performed in R (version 4.6.0).

3. Results

3.1 Carbonate Chemistry Dynamics

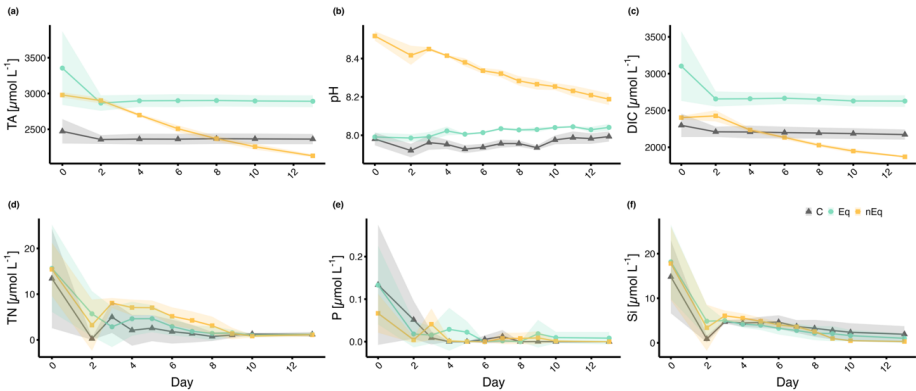


Figure 1. Changes in seawater carbonate chemistry and nutrient availability throughout the study: (a) TA, (b) pH, (c) DIC, (d) TN, (e) P, and (f) Si. Data are presented as mean ± SD (n = 3).



TA increased by approximately  $500 \pm 50 \mu\text{mol kg}^{-1}$  in both OAE treatments relative to the Control immediately following alkalinity addition (Fig. 1a). TA remained stable throughout the experiment in the Eq-OAE treatment (final TA:  $\sim 3000 \mu\text{mol kg}^{-1}$ ) and the Control (final TA:  $\sim 2480 \mu\text{mol kg}^{-1}$ ). In contrast, TA gradually declined in the nEq-OAE treatment, reaching approximately  $2380 \mu\text{mol kg}^{-1}$  by day 14, corresponding to a loss of  $\sim 20\%$  of the initial TA enhancement. Patterns in pH and DIC generally followed the temporal changes in TA (Fig. 1b, c). Both parameters remained relatively stable in the Eq-OAE and Control treatments throughout the experiment, whereas both declined progressively in the nEq-OAE treatment.

Macronutrient concentrations decreased over time irrespective of treatment (Fig. 1d, e, f). Nitrate and nitrite (TN) concentrations declined from approximately  $15$  to  $8 \mu\text{mol L}^{-1}$  within the first two days and continued to decrease gradually thereafter. Phosphate (P) decreased from  $0.10 \mu\text{mol L}^{-1}$  to below the detection limit, while dissolved silica (Si) declined from approximately  $18$  to  $5 \mu\text{mol L}^{-1}$  over the same period.

### 3.2 Plankton Biomass and Community Dynamics

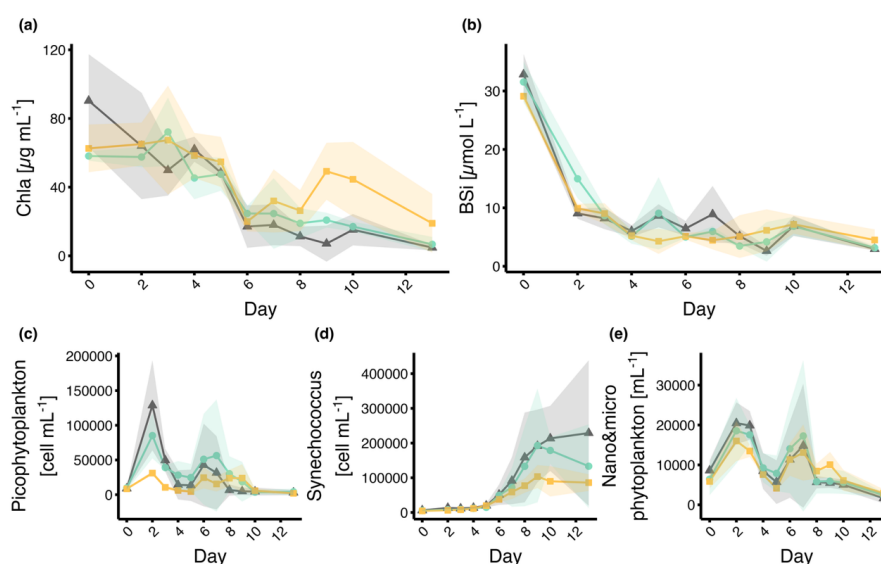


Figure 2. Phytoplankton biomass indicators and cell abundances: (a) Chla, (b) BSi, (c) picophytoplankton cell counts, (d) Synechococcus cell counts and (e) nano- and micro-phytoplankton cell counts throughout the experimental period. Data are presented as mean  $\pm$  SD ( $n = 3$ ).

Chla varied over the course of the experiment but did not differ significantly among treatments ( $p > 0.05$ ), indicating no detectable effect of either OAE treatment on phytoplankton biomass (Fig. 2a). Likewise, BSi concentrations were not significantly affected by OAE ( $p > 0.05$ , Fig. 2b). Instead, BSi exhibited a pronounced temporal decline that paralleled the depletion of dissolved nutrients. Initial BSi levels were high, likely due to resuspended particles caused by windy conditions at the time of sample collection.

Neither eukaryotic phytoplankton nor Synechococcus abundances differed significantly among treatments throughout the experiment ( $p > 0.05$ ), although both groups displayed temporal variability that was similar across all treatments (Fig. 2c, d, e).

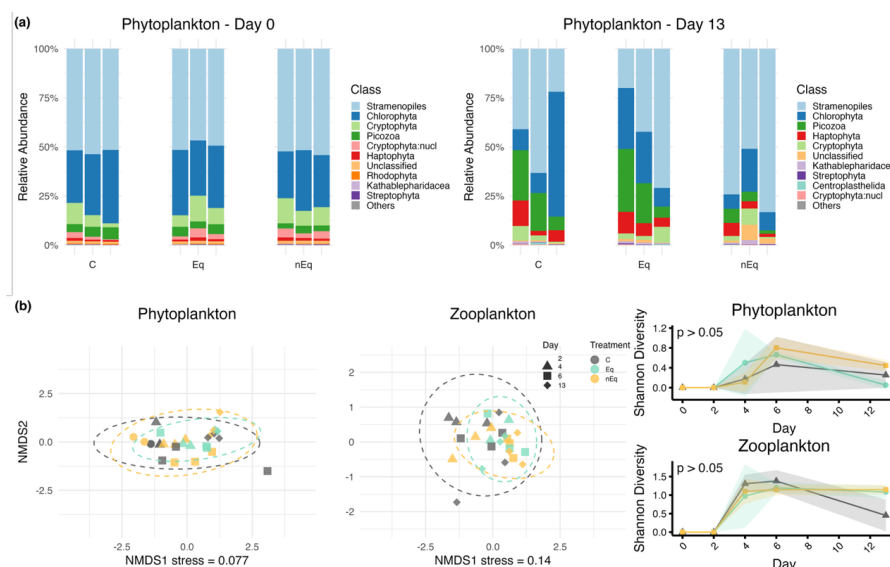


Figure 3. Plankton community composition and diversity throughout the experimental period: (a) relative abundance of major taxa at the class level based on 18S rRNA gene sequencing, (b) NMDS ordination of community structure based on microscopy data (community composition on Day 0 and 2 were not calculated due to the dominance of small-sized phytoplankton), and (c) temporal dynamics of microplankton community diversity based on microscopy data. Data are presented as mean  $\pm$  SD ( $n = 3$ ).

Consistent with Chla dynamics, neither phytoplankton nor microzooplankton abundances responded significantly to either OAE treatment throughout the experiment ( $p > 0.05$ ). Phytoplankton assemblages were dominated by small-sized cells ( $< 5 \mu\text{m}$ ) until day 4, with no larger plankton observed by microscopy. Amplicon sequencing further suggested that Chlorophyta was the predominant phytoplankton group during this period (Fig. 3a).

OAE treatments did not alter plankton community succession, as taxonomic composition followed a same temporal trajectory as the treatments, transitioning from autotroph-dominated assemblages to communities increasingly dominated by mixotrophic and heterotrophic taxa (Fig. 3b, PERMANOVA,  $p > 0.05$ ). The microzooplankton community was dominated by dinoflagellates and ciliates, with no significant treatment-induced changes in community composition. Likewise, community diversity did not differ significantly among treatments ( $p > 0.05$ ), indicating that OAE had no detectable effect on plankton biodiversity (Fig. 3c).

### 3.3 Bacterial Community Composition and Diversity

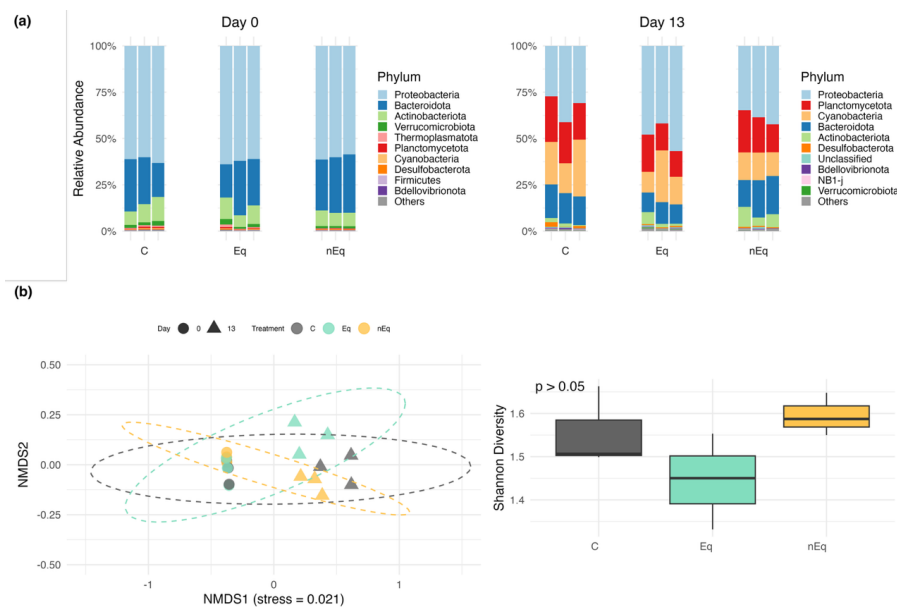


Figure 4. Bacterial community composition and alpha diversity based on 16S rRNA gene sequencing: (a) relative abundance of dominant phyla across experimental treatments, (b) NMDS ordination of bacterial community structure (Bray-Curtis dissimilarity), and (c) Shannon diversity index of the bacterial community. Data are presented as mean  $\pm$  SD ( $n = 3$ ).

NMDS ordination revealed substantial overlap among bacterial communities in the Eq-OAE, nEq-OAE, and Control treatments, with no clear separation of bacterial communities in ordination space (Fig. 4a, b). Consistent with this pattern, PERMANOVA detected no significant effect of either OAE treatment on bacterial community composition ( $p > 0.05$ ), indicating that neither OAE approach detectably altered bacterial assemblage structure over the course of the experiment. Similarly, Shannon diversity did not differ significantly among treatments ( $p > 0.05$ ), suggesting that bacterial diversity remained unaffected by OAE (Fig. 4c).

### 3.4 Particulate and Dissolved Organic Matter

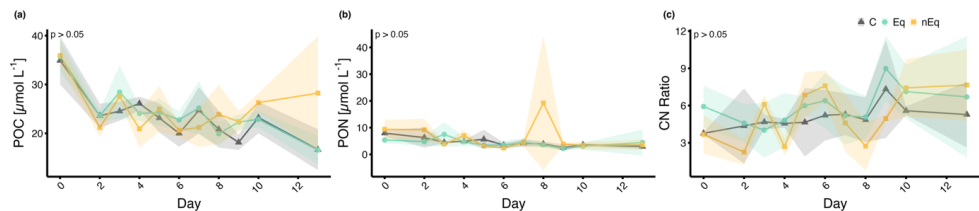


Figure 5. Temporal dynamics of particulate organic matter and stoichiometry: (a) POC concentration, (b) PON concentration, and (c) CN molar ratio throughout the experimental period. Data are presented as mean  $\pm$  SD ( $n = 3$ ).

Neither POC nor PON concentrations were significantly affected by OAE, despite temporal changes associated with plankton succession observed over the course of the experiment. The particulate CN ratio ranged between 3 and 9 and remained comparable among treatments ( $p > 0.05$ ), indicating that OAE did not influence the elemental stoichiometry of suspended particulate organic matter.

## 4. Discussion



This study evaluated the ecological and biogeochemical responses of a eutrophic NCS plankton community to OAE. Standardised methods were employed in this study, following OAEIIP-standardised protocols designed to systematically evaluate the environmental impacts of OAE across diverse marine ecosystems. Our microcosm experiment yielded two key findings: (1) neither the Eq-OAE nor the nEq-OAE treatment ( $\Delta TA \approx 500 \mu\text{mol kg}^{-1}$ ) produced detectable changes in plankton biomass, community composition, biodiversity, or particulate organic carbon, and (2) substantial TA loss occurred under the nEq-OAE treatment, indicating reduced alkalinity retention under non-equilibrated conditions. Together, these results provide insight into both the ecological responses and the practical constraints of OAE in warm coastal waters.

#### 4.1 Resilience of Plankton Community to OAE in the NCS

Across all biological variables examined, including Chla, phytoplankton and microzooplankton abundance, community composition, bacterial assemblages, biodiversity, and particulate organic matter, we detected no significant responses to either OAE treatment. Although TA in the nEq-OAE treatment gradually declined during the incubation, TA remained close to the target enhancement during the early phase of the experiment (until day 4), yet no biological responses were observed. This suggests that the carbonate chemistry perturbation imposed during this period did not produce detectable ecological effects on the plankton community.

The lack of detectable responses is likely be attributed to the limited physiological sensitivity of the resident plankton community to the carbonate chemistry perturbation imposed in this study. OAE modifies seawater carbonate chemistry by increasing pH and altering the relative abundance of dissolved  $\text{CO}_2$ ,  $\text{HCO}_3^-$  and  $\text{CO}_3^{2-}$  ions. However, physiological responses generally occur only when these changes exceed species-specific tolerance or acclimation thresholds. Previous studies have similarly shown that biological responses to OAE depend strongly on the magnitude of alkalinity enhancement, with measurable effects becoming more apparent only under substantially larger perturbations (Gately et al., 2023; Traboni et al., 2025; Xin et al., 2024). The targeted alkalinity enhancement ( $\Delta TA \sim 500 \mu\text{mol kg}^{-1}$ ) therefore appears to have remained within the physiological tolerance range of the NCS plankton community present in our study (Rost et al., 2003). This pattern may also be related to the environmental characteristics of the study region. The eutrophic coastal waters of the NCS experience pronounced fluctuations in temperature, nutrient availability, salinity, and carbonate chemistry associated with riverine inputs, coastal currents, and high biological productivity. Organisms inhabiting such variable environments may consequently possess greater physiological plasticity than those in more stable oceanic systems.

Our findings are consistent with recent OAE studies reporting minimal or no changes in natural plankton communities following moderate alkalinity enhancement (Ferderer et al., 2022; Gately et al., 2023; Marín-Samper et al., 2024; Sánchez et al., 2024; Xin et al., 2024). Similar conclusions have also been reported for higher trophic levels, including marine fishes exposed to  $\Delta TA$  of approximately  $600 \mu\text{mol kg}^{-1}$  (Goldenberg et al., 2024). Collectively, these studies suggest that moderate OAE perturbations may have limited short-term effects on many marine communities, although responses will ultimately depend on local environmental conditions, community composition, exposure duration, and the magnitude and chemical form of alkalinity addition.

It is also important to consider that, under field conditions, alkalinity-enriched seawater from point sources would be progressively diluted following release, reducing carbonate chemistry perturbations relative to those imposed in enclosed microcosms (Bach et al., 2026). Consequently, the biological responses observed here may represent a relatively strong exposure scenario compared with the transient perturbations expected following dilution in the field.

#### 4.2 Biological Carbon Sequestration Potential Under OAE

Consistent with the overall lack of detectable shifts in plankton biomass, community structure and biodiversity across experimental treatments, we observed no statistically significant differences in POC and PON among the treatments. This suggests that over the timescale examined, there was little evidence that



moderate alkalinity enhancement influenced the measured pools of particulate organic carbon and nitrogen.

270 These observations agree with previous studies reporting negligible effects of OAE on biological carbon sequestration in coastal oligotrophic systems (Paul et al., 2024; Suessle et al., 2024). The invariant POC concentrations are consistent with the absence of detectable changes in plankton biomass and community structure observed in this study. These results suggest that, under the experimental conditions tested, OAE-induced changes in inorganic carbonate chemistry were not accompanied by detectable changes in the measured particulate organic carbon pool. This apparent decoupling between the carbonate chemistry response and measured particulate organic matter highlights the potential for OAE to alter inorganic carbon cycling without necessarily producing short-term changes in plankton-derived particulate organic matter. However, longer-term experiments and measurements of carbon fixation, export, and remineralisation are needed to determine whether this pattern persists over ecologically relevant timescales.

#### 4.3 Implications for OAE

280 A notable finding observed in this study was the progressive loss of TA in the nEq-OAE treatment, which was not observed in the Eq-OAE or Control treatments. This TA loss was accompanied by a parallel decrease in DIC, yielding an approximate TA:DIC loss ratio of 2:1, indicative of calcium carbonate ( $\text{CaCO}_3$ , aragonite) precipitation. Although mineral precipitation was not directly quantified, the observed stoichiometric relationship suggests that  $\text{CaCO}_3$  formation may have contributed substantially to the observed alkalinity loss. High experimental temperatures (here 24–28 °C) likely increased carbonate mineral saturation states and promoted precipitation, which would consume both TA and DIC as  $\text{CaCO}_3$  forms (Suitner et al., 2024).

290 Additionally, the presence of suspended particles and/or biological surfaces (e.g., plankton-derived particles and detritus) in the natural seawater may have acted as nucleation sites, facilitating  $\text{CaCO}_3$  nucleation and TA loss (Hartmann et al., 2023). Similar processes have been reported in previous OAE experiments conducted under warm or highly supersaturated conditions (Hartmann et al., 2023; Moras et al., 2022; Suitner et al., 2024). The loss of TA in the nEq-OAE treatment highlights the need for careful deployment methodologies to minimise TA loss, particularly in warm coastal environments such as the NCS.

295 The stability of TA in Eq-OAE suggests that alkalised seawater bears a lower risk of TA loss after equilibrating with the atmosphere. Consequentially, equilibrating seawater with atmospheric  $\text{CO}_2$  before deployment substantially could improve alkalinity retention, although this approach also entails additional practical and economic costs, including infrastructure, operational complexity, and energy consumption. This trade-off between TA retention and implementation cost underscores the importance of optimising OAE deployment strategies for specific regional conditions—e.g., Eq-OAE may warrant consideration in 300 warm,  $\text{CaCO}_3$ -saturated coastal systems like the NCS, where TA loss risk may be high.

Several limitations should be considered when interpreting these results. First, our findings are region-specific to the eutrophic NCS and may not be generalisable to other marine systems with different plankton communities and environmental conditions. In addition, our microcosm experiment was conducted under 305 controlled laboratory conditions with limited duration, and therefore cannot fully reproduce the complex, dynamic environmental conditions of the NCS. Seasonal hydrographic variability, water-column mixing, grazing dynamics, and episodic nutrient inputs may all influence ecosystem responses under natural conditions. Second, only a single alkalinity enhancement level and one alkalinity source were investigated. Biological responses may differ under higher TA additions, alternative alkaline materials, or longer exposure periods. In addition, this study focused primarily on plankton community responses and did not evaluate 310 other potentially important ecological processes, including higher trophic levels or benthic communities. Future studies should integrate these components to develop a holistic understanding of OAE's ecological impacts in the NCS.



315 Despite these limitations, our study provides a standardised assessment of the short-term effects of OAE  
 on plankton communities in the eutrophic NCS and contributes region-specific data to the OAEPIIP  
 framework. Expanding comparable experiments across broader environmental gradients will improve  
 understanding of how ecosystem characteristics influence the ecological consequences of OAE and  
 support the development of regionally appropriate deployment strategies.

## 5. Conclusion

320 Our results demonstrate that moderate OAE ( $\Delta TA \approx 500 \mu\text{mol kg}^{-1}$ ) did not measurably affect plankton  
 biomass, community composition, biodiversity, bacterial assemblages, or particulate organic carbon in the  
 eutrophic NCS. These findings suggest that, under the conditions tested, OAE can enhance seawater  
 alkalinity without causing detectable short-term changes in the measured properties of coastal plankton  
 communities. In contrast, substantial alkalinity loss occurred under the non-equilibrated OAE treatment,  
 325 highlighting carbonate precipitation as an important consideration for OAE implementation in warm  
 coastal waters. Improving alkalinity retention through optimised deployment strategies, including  $\text{CO}_2$   
 equilibration where appropriate, will be critical for improving the efficiency of OAE. Overall, this study  
 provides evidence that moderate OAE can be implemented without detectable short-term effects on  
 productive coastal plankton communities while emphasising that deployment efficiency depends not only  
 330 on biological responses but also on the retention of added alkalinity. Future studies should investigate  
 longer-term responses, a wider range of alkalinity enhancement scenarios, and additional ecosystem  
 components to support comprehensive environmental assessments of OAE across diverse marine  
 environments.

## Data availability

335 The raw data will be made available by the authors, without undue reservation. The data will be submitted  
 to Pangaea, <https://www.pangaea.de/>.

## Author contribution

XX and JL designed the experiments. XX, YL, RC, RL, MZ and XW carried them out. XX prepared the  
 manuscript with contributions from all co-authors.

## 340 Declaration of interest

The authors declare that they have no conflict of interest.

## Acknowledgements

345 We are deeply grateful to Dr. Lennart Bach and Norfaizny Hasweera for their invaluable assistance with  
 this manuscript. A special thank goes to Dr. Shu Yang and Dr. Yuqiu Wei for their technical support with  
 sample measurement. The study was funded by the Ocean Alkalinity Enhancement Pelagic Impact  
 Intercomparison Project (OAEPIIP) and supported by the National Natural Science Foundation of China  
 (Grant No. 42606171). Xiaoke Xin was partially funded by Shandong Postdoctoral Science Foundation.

## Reference

- 350 Bach, L. T.: The additionality problem of ocean alkalinity enhancement, *Biogeosciences*, 21, 261–277,  
<https://doi.org/10.5194/bg-21-261-2024>, 2024.
- Bach, L. T., Gill, S. J., Rickaby, R. E. M., Gore, S., and Renforth, P.:  $\text{CO}_2$  Removal With Enhanced Weathering  
 and Ocean Alkalinity Enhancement: Potential Risks and Co-benefits for Marine Pelagic Ecosystems,  
*Frontiers in Climate*, 1, 7, <https://doi.org/10.3389/fclim.2019.00007>, 2019.
- 355 Bach, L. T., Ferderer, A. J., LaRoche, J., and Schulz, K. G.: Technical note: Ocean Alkalinity Enhancement  
 Pelagic Impact Intercomparison Project (OAEPIIP), *EGUsphere*, 2024, 1–22,  
<https://doi.org/10.5194/egusphere-2024-692>, 2024.



- Bach, L. T., Tyka, M. D., Wang, B., and Fennel, K.: Lethal by Design? Resolving Differences Between Experimental and Real-World Alkalinity Perturbations in Ocean Alkalinity Enhancement, *Journal of Geophysical Research: Oceans*, 131, e2025JC023598, <https://doi.org/10.1029/2025JC023598>, 2026.
- 360 Chen, S. M., Riebesell, U., Schulz, K. G., von der Esch, E., Achterberg, E. P., and Bach, L. T.: Temporal dynamics of surface ocean carbonate chemistry in response to natural and simulated upwelling events during the 2017 coastal El Niño near Callao, Peru, *Biogeosciences*, 19, 295–312, <https://doi.org/10.5194/bg-19-295-2022>, 2022.
- 365 Eggers, S. L., Lewandowska, A. M., Barcelos e Ramos, J., Blanco-Ameijeiras, S., Gallo, F., and Matthiessen, B.: Community composition has greater impact on the functioning of marine phytoplankton communities than ocean acidification, *Global Change Biology*, 20, 713–723, <https://doi.org/10.1111/gcb.12421>, 2014.
- Ferderer, A., Chase, Z., Kennedy, F., Schulz, K. G., and Bach, L. T.: Assessing the influence of ocean alkalinity enhancement on a coastal phytoplankton community, *Biogeosciences*, 19, 5375–5399, <https://doi.org/10.5194/bg-19-5375-2022>, 2022.
- 370 Gately, J. A., Kim, S. M., Jin, B., Brzezinski, M. A., and Iglesias-Rodriguez, M. D.: Coccolithophores and diatoms resilient to ocean alkalinity enhancement: A glimpse of hope?, *Science Advances*, 9, eadg6066, <https://doi.org/10.1126/sciadv.adg6066>, 2023.
- Goldenberg, S. U., Riebesell, U., Brüggemann, D., Börner, G., Sswat, M., Folkvord, A., Couret, M., Spjelkavik, S., Sánchez, N., Jaspers, C., and Moyano, M.: Early life stages of fish under ocean alkalinity enhancement in coastal plankton communities, *Biogeosciences*, 21, 4521–4532, <https://doi.org/10.5194/bg-21-4521-2024>, 2024.
- 375 Guo, J. A., Strzepek, R. F., Yuan, Z., Swadling, K. M., Townsend, A. T., Achterberg, E. P., Browning, T. J., and Bach, L. T.: Effects of ocean alkalinity enhancement on plankton in the Equatorial Pacific, *Communications Earth & Environment*, 6, 270, <https://doi.org/10.1038/s43247-025-02248-7>, 2025.
- 380 Hartmann, J., Suitner, N., Lim, C., Schneider, J., Marín-Samper, L., Arístegui, J., Renforth, P., Taucher, J., and Riebesell, U.: Stability of alkalinity in Ocean Alkalinity Enhancement (OAE) approaches – consequences for durability of CO<sub>2</sub> storage, *Biogeosciences*, 20, 781–802, <https://doi.org/10.5194/bg-20-781-2023>, 2023.
- Ilyina, T., Wolf-Gladrow, D., Munhoven, G., and Heinze, C.: Assessing the potential of calcium-based artificial ocean alkalization to mitigate rising atmospheric CO<sub>2</sub> and ocean acidification, *Geophysical Research Letters*, 40, 5909–5914, <https://doi.org/10.1002/2013gl057981>, 2013.
- 385 IPCC: Sections. In: *Climate Change 2023: Synthesis Report. Contribution of Working Groups I, II and III to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change* [Core Writing Team, H. Lee and J. Romero (eds.)], <https://doi.org/10.1017/9781017105932>, 2023.
- Köhler, P., Hartmann, J., and Wolf-Gladrow, D. A.: Geoengineering potential of artificially enhanced silicate weathering of olivine, *Proceedings of the National Academy of Sciences*, 107, 20228–20233, <https://doi.org/10.1073/pnas.1000545107>, 2010.
- Kousoulas, K., Ferderer, A., Eriksen, R., and Bach, L. T.: Winners and losers under hydroxide-based ocean alkalinity enhancement in a Tasmanian plankton community, *Journal of Phycology*, 61, 989–1006, <https://doi.org/10.1111/jpy.70052>, 2025.
- 395 Lueker, T. J., Dickson, A. G., and Keeling, C. D.: Ocean pCO<sub>2</sub> calculated from dissolved inorganic carbon, alkalinity, and equations for K<sub>1</sub> and K<sub>2</sub>: validation based on laboratory measurements of CO<sub>2</sub> in gas and



- seawater at equilibrium, *Marine Chemistry*, 70, 105–119, [https://doi.org/10.1016/s0304-4203\(00\)00022-0](https://doi.org/10.1016/s0304-4203(00)00022-0), 2000.
- 400 Marín-Samper, L., Arístegui, J., Hernández-Hernández, N., Ortiz, J., Archer, S. D., Ludwig, A., and Riebesell, U.: Assessing the impact of CO<sub>2</sub>-equilibrated ocean alkalinity enhancement on microbial metabolic rates in an oligotrophic system, *Biogeosciences*, 21, 2859–2876, <https://doi.org/10.5194/bg-21-2859-2024>, 2024.
- 405 McElhany, P., Cape, M., Faucher, G., Frieder, C., Hemery, L. G., Iglesias-Rodriguez, D., Murray, C. S., and Noble, W.: Biological thresholds for marine carbon dioxide removal (mCDR): the effect of changes in carbonate chemistry, *Biogeosciences*, 2026.
- Moras, C. A., Bach, L. T., Cyronak, T., Joannes-Boyau, R., and Schulz, K. G.: Ocean Alkalinity Enhancement – Avoiding runaway CaCO<sub>3</sub> precipitation during quick and hydrated lime dissolution, *Biogeosciences*, 3537–3557, <https://doi.org/10.5194/bg-19-3537-2022>, 2022.
- 410 Mortlock, R. A. and Froelich, P. N.: A simple method for the rapid determination of biogenic opal in pelagic marine sediments, *Deep Sea Research Part A. Oceanographic Research Papers*, 36, 1415–1426, [https://doi.org/10.1016/0198-0149\(89\)90092-7](https://doi.org/10.1016/0198-0149(89)90092-7), 1989.
- OAEIIP: Additional resources on carbonate chemistry calculation and statistical evaluations of OAEIIP, <https://doi.org/https://doi.org/10.5281/zenodo.10783751>, 2024.
- 415 Oschlies, A., Bach, L. T., Rickaby, R. E. M., Satterfield, T., Webb, R., and Gattuso, J.-P.: Climate targets, carbon dioxide removal, and the potential role of ocean alkalinity enhancement, *State of the Planet*, 2-oae2023, 1, <https://doi.org/10.5194/sp-2-oae2023-1-2023>, 2023.
- Paul, A. J., Haunost, M., Goldenberg, S. U., Hartmann, J., Sánchez, N., Schneider, J., Suitner, N., and Riebesell, U.: Ocean alkalinity enhancement in an open ocean ecosystem: Biogeochemical responses and carbon storage durability, *EGUsphere*, [preprint], <https://doi.org/10.5194/egusphere-2024-417>, 2024.
- 420 Renforth, P. and Henderson, G.: Assessing ocean alkalinity for carbon sequestration, *Reviews of Geophysics*, 55, 636–674, <https://doi.org/10.1002/2016RG000533>, 2017.
- Rost, B., Riebesell, U., Burkhardt, S., and Sültemeyer, D.: Carbon acquisition of bloom-forming marine phytoplankton, *Limnology and oceanography*, 48, 55–67, <https://doi.org/10.4319/lo.2003.48.1.0055>, 2003.
- 425 Sánchez, N., Goldenberg, S. U., Brüggemann, D., Jaspers, C., Taucher, J., and Riebesell, U.: Plankton food web structure and productivity under ocean alkalinity enhancement, *Science Advances*, 10, eado0264, <https://doi.org/10.1126/sciadv.ado0264>, 2024.
- Schulz, K. G., Bach, L. T., and Dickson, A. G.: Seawater carbonate system considerations for ocean alkalinity enhancement research, *State of the Planet Discussions*, 2023, 1–24, <https://doi.org/10.5194/sp-2023-12>, 2023.
- 430 Subhas, A. V., Marx, L., Reynolds, S., Flohr, A., Mawji, E. W., Brown, P. J., and Cael, B. B.: Microbial ecosystem responses to alkalinity enhancement in the North Atlantic Subtropical Gyre, *Frontiers in Climate*, 4, <https://doi.org/10.3389/fclim.2022.784997>, 2022.
- 435 Suessle, P., Taucher, J., Goldenberg, S. U., Baumann, M., Spilling, K., Noche-Ferreira, A., Vanharanta, M., and Riebesell, U.: Particle fluxes by subtropical pelagic communities under ocean alkalinity enhancement, *Biogeosciences*, 22, 71–86, <https://doi.org/10.5194/bg-22-71-2025>, 2024.



- Suitner, N., Faucher, G., Lim, C., Schneider, J., Moras, C. A., Riebesell, U., and Hartmann, J.: Ocean alkalinity enhancement approaches and the predictability of runaway precipitation processes: results of an experimental study to determine critical alkalinity ranges for safe and sustainable application scenarios, *Biogeosciences*, 21, 4587–4604, <https://doi.org/10.5194/egusphere-2023-2611>, 2024.
- 440 Tak, Y.-J., Cho, Y.-K., Hwang, J., and Kim, Y.-Y.: Frontiers | Assessments of Nitrate Budgets in the Yellow Sea Based on a 3D Physical-Biogeochemical Coupled Model, *Frontiers in Marine Science*, 8, <https://doi.org/10.3389/fmars.2021.785377>, 2022.
- Traboni, C., Nocera, A. C., Romano, F., Courboulès, J., Chantzaras, C., Magiopoulos, I., Varliero, S., Basso, D., and Pitta, P.: Plankton do not care: Minimal effects of ocean liming on plankton growth and grazing in the Eastern Mediterranean, *Limnology and Oceanography*, <https://doi.org/10.1002/lno.70136>, 2025.
- 445 Utermöhl, H.: Zur vervollkommnung der quantitativen phytoplankton-methodik: Mit 1 Tabelle und 15 abbildungen im Text und auf 1 Tafel, *Internationale Vereinigung für theoretische und angewandte Limnologie: Mitteilungen*, 9, 1–38, <https://doi.org/10.1080/05384680.1958.11904091>, 1958.
- Wang, Y., Dou, Y., Zou, L., Gao, F., Su, D., Hu, R., Yue, B., and Xue, B.: Summery Intra-Tidal Variations of Suspended Sediment Transportation – Topographical Response and Dynamical Mechanism in the Aoshan Bay and Surrounding Area, Shandong Peninsula, *J. Ocean Univ. China*, 20, 1398–1408, <https://doi.org/10.1007/s11802-021-4656-9>, 2021.
- 450 Xin, X., Goldenberg, S. U., Taucher, J., Stühr, A., Arístegui, J., and Riebesell, U.: Resilience of Phytoplankton and Microzooplankton Communities under Ocean Alkalinity Enhancement in the Oligotrophic Ocean, *Environmental Science & Technology*, 58, 20918–20930, <https://doi.org/10.1021/acs.est.4c09838>, 2024.
- 455