Nueces River Tidal Nutrient Bioassays

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Executive summary

Frequent, severe fish kills in the Nueces River's tidal segment have raised concerns about deteriorating water quality. Previous research to investigate these fish kills linked mortality to low dissolved oxygen events, which may be related to phytoplankton blooms that are indicative of excessive nutrient loadings. These issues were noted in the 2022 TCEQ Texas Integrated Report. However, there is little data available to decision-makers and resource managers regarding the nutrient(s) that supports phytoplankton growth, information necessary for formulating strategies to address water quality degradation in the system. To determine whether nitrogen or phosphorus controls phytoplankton growth in the system, nutrient bioassays were conducted in winter, early summer, late summer and fall 2024 in the Nueces River tidal segment. Three of the four experiments showed a definitive phytoplankton growth response to nitrogen addition relative to the control (ambient nutrient levels) and phosphorus addition treatments. A fourth experiment showed a slightly higher, but statistically indistinct growth response to nitrogen + phosphorus addition compared to nitrogen addition alone, but the response to both treatments was much higher than in the control and phosphorus addition treatments. These findings suggest that nitrogen load management should be a priority for mitigating eutrophication and water quality degradation in the system. Interestingly, the effects of nutrient availability appeared to vary with season/temperature, with a stronger growth response observed during cooler months of the year. This suggests that there may be a crucial window (i.e., cooler months) for nutrient management in the system, similar to what has been observed in other estuarine environments. Additional monitoring and experimentation is recommended to determine potential seasonality in terms of nutrient regulation of phytoplankton growth in the Nueces River tidal segment.

Acknowledgement

We extend our heartfelt gratitude to the staff and students who contributed to the experiment and field sampling efforts; their dedication and hard work were instrumental in the success of this project. We are extremely thankful to Mr. Jon Schroeder for allowing us to set up the experiment and provide additional logistical support on his property. Our deepest appreciation also goes to the Coastal Bend Bays & Estuaries Program for providing the necessary funds and resources to make this work possible.

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Introduction

Increasing frequency and severity of fish kills within the tidal segment of the Nueces River have raised concerns about water quality degradation. Previous research to investigate these fish kills linked mortality primarily to low dissolved oxygen events (unpubl. TPWD reports). The 2022 TCEQ Texas Integrated Report also noted a concern level for chlorophyll *a*, an indicator of phytoplankton biomass. High phytoplankton biomass can trigger low oxygen conditions in estuarine systems similar to the Nueces River tidal segment (NRC 2000).

High and/or increasing phytoplankton biomass is often indicative of excessive nutrient loadings to a waterbody (NRC 2000). Possible sources of excess nutrient loadings to the Nueces River tidal segment include permitted point source discharges, runoff from urban and rural non-point sources, and septic tanks, among other sources. In addition, the Nueces River tidal has a median residence time of >4 months due to a lack of tidal flushing and freshwater inflow (Wetz unpubl. data), which can exacerbate symptoms of excessive nutrient loadings (NRC 2000; Bricker *et al*. 2008).

Concerns about the health of the Nueces River tidal segment have been expressed by stakeholders and led to the formation of the Nueces River Tidal Stakeholder Group in 2023. However, little information is available on possible nutrient sources and drivers of water quality degradation in it. The ***goal*** of the present study was to identify the main nutrient(s) controlling phytoplankton growth in the Nueces River tidal segment.

Methods

Experimental design

The study employed nutrient addition bioassay experiments to evaluate the role of nutrients in controlling phytoplankton growth. Experiments were conducted during winter (27th to 29th February 2024), early summer (4th to 6th June 2024), late summer (13th to 15th August 2024), and fall (20th to 22nd November 2024). Experimental water was collected from the Nueces River tidal segment (27.886119 N, -97.609558 E), in the vicinity of where symptoms of water quality degradation are most pronounced (Fig. 1). Water was collected in two separate 20-L acid-washed, deionized water-rinsed carboys from 0.1 m depth.

Experiments were conducted by incubating water exposed to various treatments in 4-L polycarbonate Cubitainers that were transparent to sunlight. Treatments (in triplicate) included: nitrogen (20 µM N addition as NH_4Cl; "+N"), phosphorus (2 µM P addition as NaH_2PO_4; "+P"), N+ P (20 µM N addition as NH_4Cl, 2 µM P addition as NaH_2PO_4), and a control

treatment with ambient nutrient concentrations (Fig. 2). Experimental Cubitainers were incubated in a 1.0 m wide x 0.5 m deep limnocorral (Fig. 3a) with one layer of neutral density mesh screen on top. The limnocorral was placed in the river at a location that was ~1 meter deep with well-flowing water and no natural shading (Fig. 3b). At the start of each experiment, physicochemical parameters including water temperature, salinity, and dissolved oxygen (DO) were measured with a handheld YSI ProPlus multiparameter sonde. To assure data quality, the probes of the multiparameter sonde were calibrated before and after the fieldwork for all measured parameters. During experiments, two HOBO® sensors were deployed in the limnocorral for measurement of light and temperature every 5 minutes (Fig. 3c).

At the start of each experiment, samples (50 ml) for chlorophyll *a* (Chl *a*) were collected from all the Cubitainers. Then the headspace was carefully removed and the Cubitainers were transferred into the limnocorral (Fig. 3d). At 24- and 48-hour time points, Cubitainers were briefly removed from the limnocorral, gently mixed to homogenize water, and water samples were drawn for Chl *a* which is a proxy for phytoplankton biomass. Water samples were stored on ice and taken back to the lab for filtration and further processing. The water samples (25 ml) were filtered using Whatman GF/F filters (pore size 0.7 µm, 25 mm diameter) and stored in vacutainers at -20°C until fluorometric analysis. The filters were placed in 90% acetone for 20-24 hrs. in the dark at 0 °C to extract Chl *a* and quantified fluorometrically using a Turner Trilogy fluorometer (Model 7200, USA) without acidification.

Results and Discussion

Experiment I (February 2024):

At the time of experimental water collection from the field, experiment #1 had the lowest water temperature (17.8°C) and salinity (7.16) of all experiments (Fig. 4). Dissolved oxygen (DO) was at its maximum (8.89 mg L^{-1}) with an oxygen saturation of 101.1%. The Secchi depth was measured at 0.2 m, while the total water depth was 1 m.

By the time the experiment started, water temperature had warmed to ~25ºC (Fig. 5). Over the course of the experiment, water temperature ranged between 13.1°C and 25.1°C, with an average of 21.5 ± 2.5°C. The water temperature gradually cooled throughout the experiment, with a sharper drop observed on the last night (Fig. 5). Daytime light intensity reached 19,814 Lux, with an average of 2,959 ± 2,933 Lux (Fig. 5). Light levels were lowest, on average, of all experiments.

The initial Chl *a* concentration was high (45 µg/L) relative to other experiments, likely due to increased nutrient availability during this period. The relatively low salinity observed at the

start of the experiment is indicative of a freshwater pulse, which in other estuaries would typically be accompanied by increased nutrient availability. Additional work is needed to determine if this is the case and if the high Chl *a* is typical of wintertime conditions in the system. Knowledge gained from a complimentary monthly monitoring program being conducted by the Center for Coastal Studies will be beneficial for this purpose.

The +N treatment led to substantially increased Chl *a* concentration, reaching 78 µg/L in 24 hours (Fig. 6). The N+P treatment followed a similar pattern to the +N treatment. The +P treatment had no effect, and the trend was comparable to that of the control group, which remained unchanged for 24 hours and then slightly declined (to 40 µg/L) after 48 hours. This experiment highlights the strong dependence of Chl *a* on nitrogen, rather than phosphate, during the winter, and the overall importance of nutrient availability in controlling phytoplankton growth under these conditions.

Experiment II (June 2024):

Experiment #2 captured the effects of early summer conditions, with higher water temperature (29.7°C) and salinity (31.15) (Fig. 4) driven by decreased inflows and increased evaporation relative to precipitation. DO concentration and oxygen saturation dropped to 3.52 mg L^{-1} and 55.8%, respectively. The Secchi depth increased slightly to 0.3 m, and the total depth was 1.4 m.

During the experiment, water temperature showed clear day/night cycles and ranged from 24.9°C to 35.6°C (Fig. 5). A sharp decrease was observed on the last night of the experiment. Light intensity was higher than during experiment #1, reaching a peak of 29,030 Lux and averaging 7,697 ± 6,591 Lux (Fig. 5).

Chl *a* was 18.9 µg/L at the start of the experiment, lower than at the beginning of experiment #1 (Fig. 4). This is possibly due to lower nutrient availability, as indicated by the high salinities that would indicate a lack of recent freshwater input and nutrient loading. Chl *a* concentration increased to 20.4 µg/L over the first 24 hr of the +N treatment, after which it declined slightly (19.9 µg/L) (Fig. 6). The N+P treatment followed a similar pattern as the +N treatment, but it continued to increase after 48 hours. In contrast, the control and +P treatments showed a substantial decrease in Chl *a* throughout the experiment (Fig. 6). In general, this experiment's findings indicate that nitrogen and/or nitrogen+phosphorus combined are important for controlling phytoplankton growth under these conditions.

Experiment III (August 2024):

At the start of experiment #3, the water temperature was high (31.5°C) but salinity was low (9.03) (Fig. 4). DO levels were very low at 2.01 mg L^{-1} and oxygen saturation was 29%. The Secchi depth remained at 0.3 m, and the total water depth was 1 m.

Over the course of the experiment, a clear day/night cycle was observed in water temperature, with lower temperatures at night and higher temperatures during the day. Water temperature ranged from 28.8°C to 36.5°C, with an average of 32.3 ± 1.8°C (Fig. 5). The highest level of light intensity recorded was 44,667 Lux, with an average of 12,408 ± 12,441 Lux.

Chl *a* was 10.2 µg/L at the start of the experiment, lower than at the start of experiment #1 or #2 (Fig. 4). Chl *a* concentration decreased in the +N and N+P treatments, reaching 6.3 µg/L at the end (Fig. 6). The control as well as +P treatments also decreased, and at a sharper rate than the other two treatments, reaching 3.8 µg/L at the end. These findings suggest that nitrogen is still the limiting nutrient for phytoplankton growth, but that top-down control, such as zooplankton grazing, might be an important factor regulating phytoplankton biomass under these conditions. Grazing in particular may also explain the relatively low Chl *a* observed at the start of the experiments.

*Experiment IV (November 2024)**:***

Water temperature was 22.8°C and salinity was 28.19 at the start of the experiment (Fig. 4). DO concentration was 3.01 mg L^{-1}, with oxygen saturation of 42.8% (Fig. 4). The Secchi depth was the highest among all experiments at 0.6 m, with a total depth of 1.3 m.

Over the course of the experiment, water temperature displayed day/night variability and ranged from 17.3°C to 27.1°C, with an average of 21.8 ± 2.8°C (Fig. 5). Light intensity averaged 14,776 ± 10,162 Lux, highest of all the experiments.

Chl *a* was 6.0 µg/L at the start of the experiment, the lowest of all the experiments (Fig. 4). For the first 24 hours, Chl *a* remained relatively stable for all treatments (Fig. 6). However, the +N and N+P treatments exhibited a 60% increase in Chl *a* concentration between 24 and 48 hours, reaching 9.2 µg/L and 9.6 µg/L, respectively. In contrast, Chl *a* remained stable in the +P and control treatments, once again indicative of nitrogen limitation of phytoplankton growth.

Management Recommendations

First and foremost, across all experiments, nitrogen was observed to have a greater influence on Chl *a* concentration compared to phosphorus. This suggests that nitrogen is the

primary limiting nutrient driving phytoplankton growth in this system, while phosphorus played a secondary role. The differential impact of nitrogen compared to phosphorus highlights the importance of nutrient management strategies focused on nitrogen control to address excessive phytoplankton growth.

Another important finding from this study, which requires further verification from dedicated monitoring that is currently underway in the Nueces River tidal segment, is the apparent seasonality in phytoplankton biomass and controls on phytoplankton growth. For example, the highest Chl *a* concentration was observed in late February. Late winter/early spring blooms are a common feature of many estuaries, and in coastal systems such as Chesapeake Bay or the Northern Gulf of Mexico, the biomass produced during these events can not only support nutrient recycling during warmer months but also contribute to oxygen declines as bacteria break down the biomass in warming conditions (Justic *et al*. 1993; Malone *et al*. 1996). Although further information is needed from dedicated monitoring to determine if this phenomenon holds true, the implication is that nitrogen management strategies may need to target the critical time period for phytoplankton biomass accumulation in the system, namely late fall through late winter/spring when phytoplankton growth response to nitrogen additions was most pronounced here.

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Figures

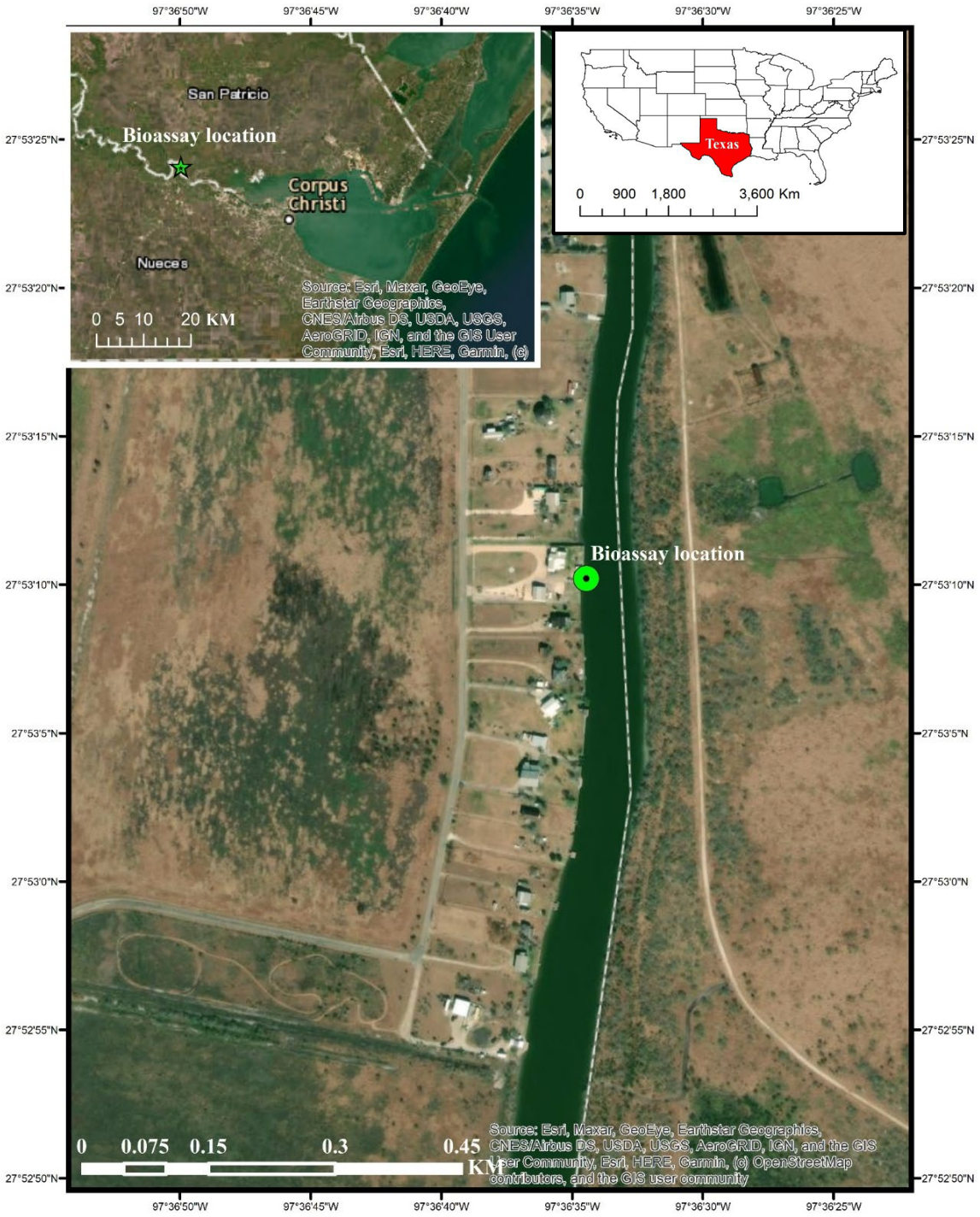

Figure 1. Location of the experiment site on the Nueces River, Corpus Christi, Texas.

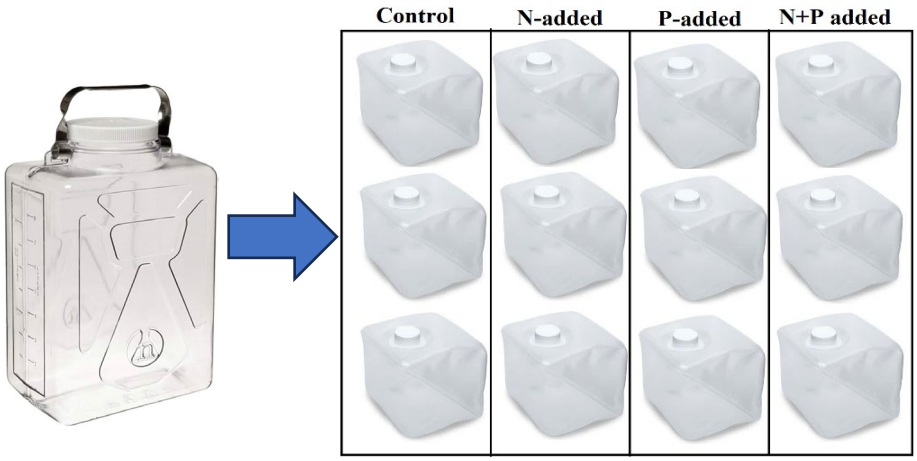

Figure 2: Four treatments were prepared (in triplicate) in each experiment: Nitrogen-added (20 µM N as NH_4Cl), Phosphorus-added (2 µM P as NaH_2PO_4), N + P added (20 µM N as NH_4Cl, 2 µM P as NaH_2PO4), and a control treatment with ambient nutrient concentrations.

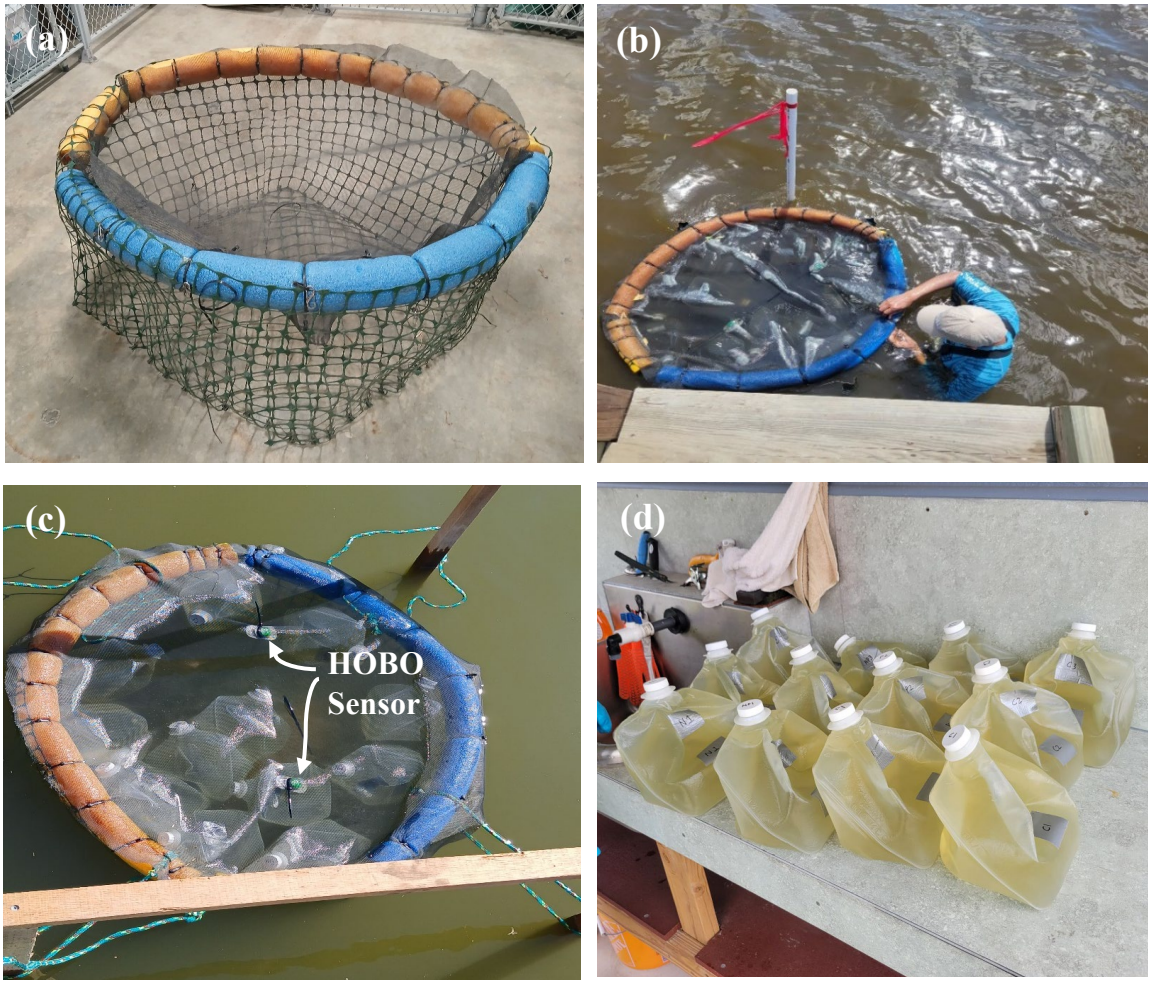

Figure 3: a) Limnocorral setup for the experiment, b) limnocorral deployed in the water, c) HOBO sensors in the limnocorral, and d) Cubitainers after collection of initial samples and removing headspace.

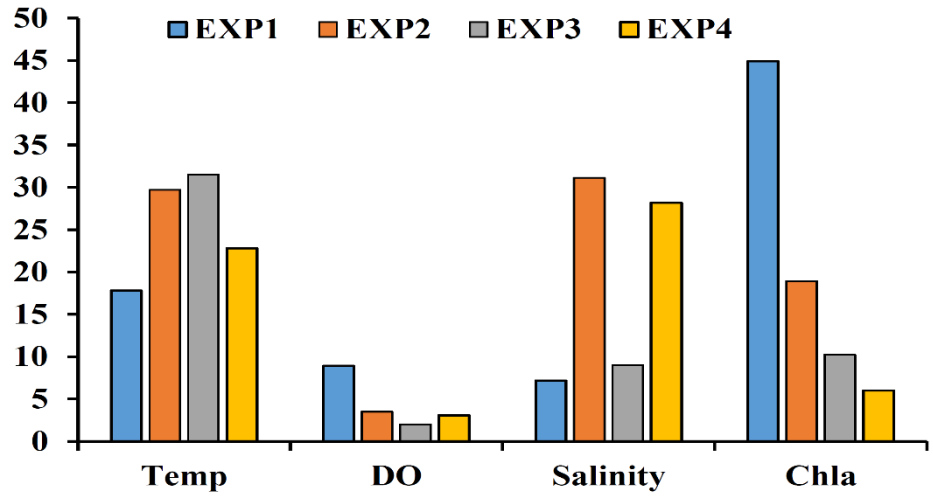

Figure 4: Physicochemical parameters of the site water used to initiate each experiment.

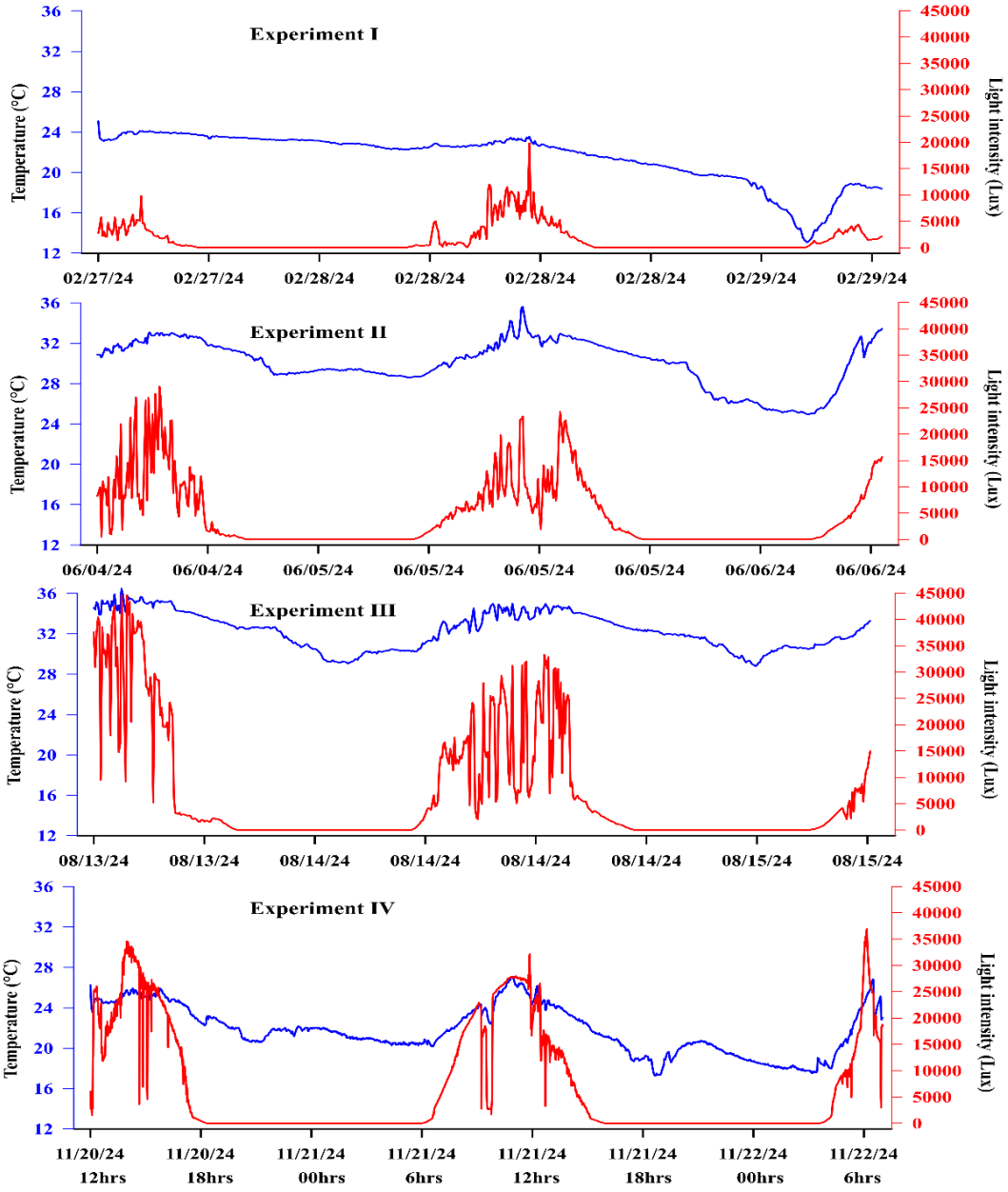

Figure 5: HOBO data of water temperature and light levels during all four experiments.

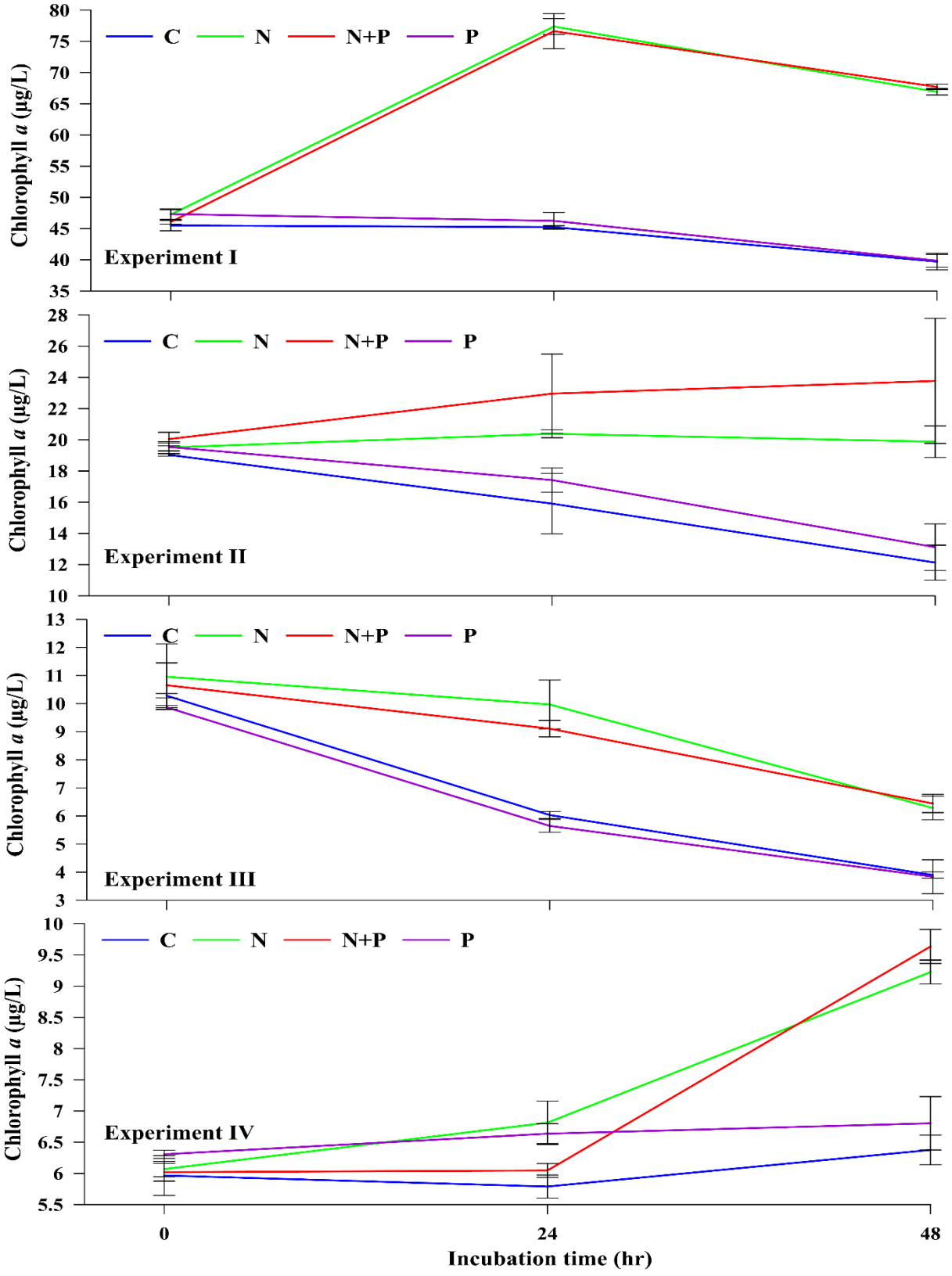

Figure 6: Changes in the chlorophyll *a* concentration of different treatments for all experiments from 0 hr. to 48 hr. Note that the range of chlorophyll concentrations is different for each experiment.