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# Relationship between water quality and phytoplankton distribution of aquaculture areas in a tropical lagoon

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**Abstract** Aquaculture activities can affect water quality and phytoplankton composition. Our study estimated phytoplankton density and composition relating to aquaculture-impacted environmental factors. We analyzed water quality and phytoplankton at 35 sites in a tropical brackish lagoon, including inside aquaculture ponds (integrated farming of fish, shrimp, and crab), at wastewater discharge points, within 300 m of these points, and farther out in the

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Faculty of Biology, University of Sciences, Hue University, Hue, Vietnam e-mail: lquangdoc@hueuni.edu.vn lagoon. Measurements were taken after aquaculture activities started in March and again in July. In both periods, total nitrogen (TN), total phosphorus (TP), chlorophyll-a (Chl-a), and turbidity decreased from the aquaculture ponds to the farther lagoon areas. Principal component analysis showed that nutrients, turbidity, and Chl-a were critical factors in aquaculture ponds, while salinity, temperature, pH, dissolved oxygen (DO), and water depth influenced water quality outside the ponds. Phytoplankton density was higher in July than in March due to aquaculture characteristics. Redundancy analysis indicated that

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phytoplankton, typical of inorganic, turbid, shallow lakes, was present throughout, whereas marine phytoplankton characterized the open water area (OWA). Marine phytoplankton caused a higher Shannon-Wiener index in July compared to March for OWA. Phytoplankton in aquaculture ponds was dominated by Oscillatoria spp., while Thalassiosira spp. dominated outside the ponds. We also identified indicator genera for two connected lagoons. Although constant water exchange prevented identifying specific indicator phytoplankton groups for aquaculture, this revealed the impact of wastewater from aquaculture ponds on the natural environment in the lagoons. Research on phytoplankton communities is necessary for the sustainable development of aquaculture and environmental management in coastal lagoons.

**Keywords** Aquaculture · Tropical lagoon · Nutrients · Phytoplankton · Functional groups · Viet Nam

### Introduction

The growth and distribution of phytoplankton that play a foundational role within the ecological food web by contributing to the accumulation and transformation of energy and matter (Rajkumar et al., 2009) are influenced by environmental factors. Their suspended existence within the aquatic environment and direct utilization of nutritional resources in water for growth and development render them to be influenced directly by environmental conditions (Reynolds, 2006; Suthers et al., 2019). Environmental factors such as temperature, light, turbidity, nutrient concentration, and filterfeeding animal communities substantially influence the distribution of phytoplankton communities (Thangaradjou et al., 2012; Spilling et al., 2015). Salinity is an important factor governing the phytoplankton community in brackish or coastal areas. Thereby, phytoplankton is frequently employed as a crucial indicator for monitoring alterations in environmental parameters and the water quality of aquatic ecosystems, primarily owing to its heightened sensitivity and swift responsiveness to shifts in environmental conditions (Fai et al., 2023; Ismail & El Zokm, 2023; Ismail et al., 2023; Reynolds et al., 1993; Seddon, 1972; Spence, 1967). Additionally, Bartozek et al. (2014) suggested that seasonal fluctuations and hydrodynamics significantly drove variations observed in phytoplankton communities and physicochemical indicators within the aquatic environment.

In aquaculture, nitrogen (N) and phosphorus (P) from residual food and fish waste in both the water column (Dunne et al., 2021; Guo et al., 2009) and the sediment (Boyd et al., 2007; Moncada et al., 2019) impact on both natural environmental quality and phytoplankton communities. Eutrophication occurs in aquaculture areas (Kang et al., 2021; Moncada et al., 2019), impacting the receiving ecosystem. According to the findings of Miranda et al. (2016), Rosini et al. (2016), and Ge et al. (2023), aquaculture has been proposed as a direct factor influencing the structural composition of phytoplankton communities.

In the Tam Giang–Cau Hai, the largest brackish lagoon system in Southeast Asia, although aquaculture production is the most important economic activity, this activity has adversely affected the lagoon's natural environment. According to Cao et al. (2013), the loads of chemical oxygen demand (COD), biochemical oxygen demand (BOD), total nitrogen (TN), and total phosphorus (TP) from aquaculture waste in 2010 in this lagoon system were 153 tons, 44 tons, 28 tons, and 25 tons, respectively. Untreated waste from aquaculture was discharged directly into the Tam Giang–Cau Hai lagoon system. Additionally, these loads were predicted to increase three times by 2020 (Cao et al., 2013).

Historically, the research in the Tam Giang–Cau Hai lagoon system was predominantly conducted to evaluate the environmental quality of the lagoon area in general (Truong et al., 2015; Nhu Y et al., 2019; Truong & Nguyen, 2020) as well as to focus on the biodiversity of microalgae, diatoms, mangroves and fish (Ton, 2009), seagrasses (Phan et al., 2017), and other submerged aquatic vegetation (Phan et al., 2018). Our study aims to evaluate the relationship between the density and composition of phytoplankton and the temporal and spatial fluctuation of environmental factors under the impact of aquaculture activities inside the ponds and within the immediate surroundings.

#### Materials and methods

Study area and sample collection

The Tam Giang–Cau Hai lagoon system is remarkable for its rich biodiversity and diverse ecosystems.

According to a report from the Center for Coastal Management and Sustainable Development, Hue University of Sciences (CMD, 2020), there were 1194 species of phytoplankton, zooplankton, benthos, fishes, amphibians, reptiles, birds, mangrove trees, and submerged aquatic vegetation, including seagrasses and seaweeds. They occur in ecosystems representing specific types of estuaries and coastal lagoons. Therefore, the Tam Giang-Cau Hai lagoon system is essential in ensuring an optimal habitat and living environment for both aquatic and terrestrial species. In 2020, the Provincial People 's Committee of Thua Thien Hue enacted Decision No. 495/ QD-UBND dated February 20, 2020, establishing the Tam Giang-Cau Hai lagoon as a Wetland Protected Area. The establishment of the Tam Giang-Cau Hai Wetland Protected Area is highly significant for the conservation and development of a typical and vital tropical marine ecosystem in Vietnam and will be an opportunity to carry out conservation activities, protect natural resources, ensure ecological balance, and maintain the natural landscape and biodiversity value for the region's long-term socio-economic development.

The research area of this study focuses on the Thuy Tu and Cau Hai lagoons in the Tam Giang-Cau Hai lagoon system, with latitude 16°14' to 16°25' North and longitude 107°38'-107°93' East. At Thuy Tu lagoon, both shores and the central water area accommodate aquaculture ponds. In the context of the Cau Hai lagoon within our study, the sampling area encompasses the connecting section between Thuy Tu and Cau Hai lagoons, to southern portions of the Cau Hai lagoon in Loc Tri Commune, Phu Loc District, and also focuses on the region situated in Giang Hai Commune-northeast of Cau Hai lagoon where aquaculture activities are concentrated. Furthermore, the western part of Cau Hai lagoon receives water from significant rivers (Dai Giang and Truoi rivers) that flow into it before discharging into the sea (Fig. 1).

Samples were collected in March and July 2021 from 35 sites with increasing distances from the aquaculture ponds (AP) and wastewater discharge points (WWDP) to points farther away, situated from 50 to 300 m from WWDP ( $\leq$  300 m) and  $\geq$  400 m from WWDP (open water area (OWA)) (Fig. 1 and Table 1). This approach aimed to assess the extent of the impact of aquaculture on the

lagoon area. The sampling details at those sites are as follows: (i) Six samples at AP (three ones in the Thuy Tu lagoon and three ones in Cau Hai lagoon); (ii) six points at WWDP (TT1, TT6, and TT10 in the Thuy Tu lagoon; CH2, CH4, and CH14 in Cau Hai lagoon); (iii) 17 points at a distance of  $\leq$  300 m (TT2, TT3, TT4, TT5, TT7, TT8, TT9, TT11, TT12, TT13, and TT14 in the Thuy Tu lagoon and CH1, CH3, CH5, CH6, CH13, and CH15 in Cau Hai lagoon); and (iv) six points at OWA including from CH7 to CH12 in the Cau Hai lagoon.

AP, aquaculture ponds; WWDP, wastewater discharge points;  $\leq 300$  m, at a distance of 50–300 m from the discharge point; OWA, open water area.

А horizontal water sampler (Model 1120-G45, USA) was used to collect samples for water quality determination and quantitative analysis of phytoplankton. Depending on the depth of each sampling location, samples were taken at the surface layer (approximately 0.3 to 0.5 m from the surface), or composite samples were collected at depths spaced 0.5 m. These samples were preserved below 4 °C in 1.5-L PET plastic bottles and then transported to the laboratory. The samples for chlorophyll-a analysis were stored in 1.5-L dark plastic bottles and analyzed within 24 h after collection.

Phytoplankton samples for qualitative analysis were collected using a conical mesh net (mesh size,  $20 \ \mu\text{m}$ ; mouth diameter of the net,  $20 \ \text{cm}$ ). This was achieved by repeatedly casting and towing the net around the sampling point approximately 7 to 10 times during the survey. The samples obtained were preserved in 150-mL plastic bottles. Samples were fixed by adding 3 mL of Lugol's solution for quantitative samples and 0.3 mL for qualitative samples. These samples were then stored at room temperature in the dark until they were analyzed in the laboratory.

Dissolved oxygen (DO), pH, salinity, temperature, and turbidity were measured by a multiple parameters device with specific sensors (HORIBA U5000, Horiba, Japan) on-site during each field trip. The water depth was measured by a depth sounder (Hondex PS-7, Honda Electronics, Japan). Total nitrogen (TN) and Chl-a concentrations were analyzed based on standard methods of APHA (2017). Total phosphorus (TP) concentration was analyzed according to the method of Menzel and Corwin (1965).

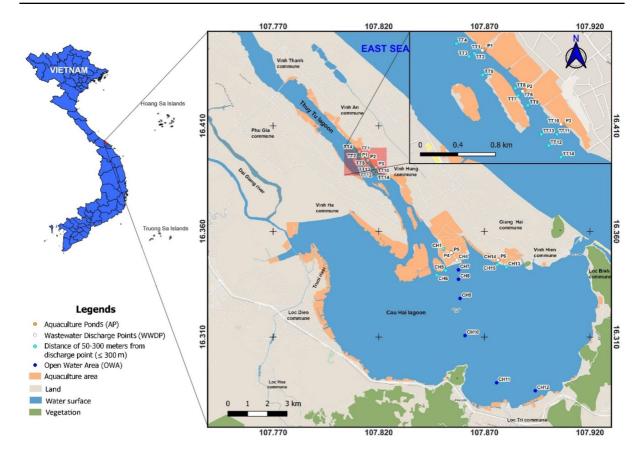


Fig. 1 Map of sampling locations in aquaculture ponds (P), Thuy Tu (TT) lagoon, and Cau Hai (CH) lagoon

Qualitative and quantitative analysis of phytoplankton

Regarding the qualitative analysis, the phytoplankton genera were determined by a morphological method with the used taxonomic keys. This identification was performed using an Olympus CX33 optical microscope at a magnification of  $\times 400$  according to the reference works of Shirota (1966), Tomas (1997), and Ton (2009) and updated according to the website https://www.algaebase.org (Guiry & Guiry, 2022).

Additionally, we classified phytoplankton into functional groups (FGs) based on the guidelines established by Reynolds et al. (2002) and updated by Padisák et al. (2009). However, these references primarily apply to freshwater environments, such as rivers and lakes. In contrast, our study area is in a brackish environment with an average salinity of 15% and even reached up to 28% at certain times and locations. Consequently, we reclassified them into "practical" phytoplankton functional groups based on the environmental characteristics of the study area (Supplementary material). Applying FGs aims to assess better the aspects of phytoplankton distribution under the impact of environmental factors.

For quantitative analysis, the initial water sample was thoroughly mixed and transferred into a 1000-mL volumetric cylinder for sedimentation and gradual water separation through multiple stages. Each sedimentation stage lasted for 24-48 h and involved a volumetric cylinder with volumes of 1000 mL, 500 mL, 200 mL, and 100 mL consecutively. Eventually, the upper portion of the water in the 100-mL volumetric cylinder was removed, and the depositing part was kept, with an approximate volume from 30 to 50 mL, for quantitative analysis. The Sedgewick-Rafter counting chamber with a volume of 1000 µL was utilized to enumerate phytoplankton calculated as the number of cells/L (Hallegraeff et al., 2003; Sournia, 1978). To reduce the absolute limits of expectations for the number of cells in each sample, the number of

 Table 1
 Location details of 35 sampling sites in aquaculture ponds (P), Thuy Tu (TT) lagoon, and Cau Hai (CH) lagoon

No	Site	Latitude	Longitude	Distance
1	P1	16.39786	107.8112	AP
2	P2	16.39382	107.8151	AP
3	P3	16.3904	107.819	AP
4	P4	16.35017	107.8543	AP
5	P5	16.34988	107.8546	AP
6	P6	16.34673	107.8768	AP
7	TT1	16.39775	107.8109	WWDP
8	TT6	16.39367	107.8149	WWDP
9	TT10	16.39039	107.8187	WWDP
10	CH2	16.34989	107.8544	WWDP
11	CH4	16.34653	107.857	WWDP
12	CH14	16.34647	107.8764	WWDP
13	TT2	16.39744	107.8101	≤300 m
14	TT3	16.39711	107.8096	≤300 m
15	TT4	16.39842	107.8083	≤300 m
16	TT5	16.39531	107.8109	≤300 m
17	TT7	16.39322	107.8145	≤300 m
18	TT8	16.39394	107.8142	≤300 m
19	TT9	16.39225	107.8154	≤300 m
20	TT11	16.38939	107.8183	≤300 m
21	TT12	16.38825	107.8175	≤300 m
22	TT13	16.38939	107.8168	≤300 m
23	TT14	16.38711	107.8187	≤300 m
24	CH1	16.35172	107.8507	≤300 m
25	CH3	16.34972	107.8542	≤300 m
26	CH5	16.34153	107.8516	≤300 m
27	CH6	16.33931	107.8474	≤300 m
28	CH13	16.34308	107.8804	≤300 m
29	CH15	16.34425	107.8761	≤300 m
30	CH7	16.34181	107.8577	OWA
31	CH8	16.33736	107.8578	OWA
32	CH9	16.32828	107.8585	OWA
33	CH10	16.31069	107.8608	OWA
34	CH11	16.28836	107.8758	OWA
35	CH12	16.28456	107.8941	OWA

cells was counted without repetition within a range of 400 to 500 cells per sample of each genus.

#### Data analysis

The Kruskal–Wallis test (a one-way ANOVA for nonparametric data) was employed to assess the differences among various environmental factors and phytoplankton cell density from within the pond to the outside area of open water in the lagoon by different distances and the monitoring moments (March and July).

To assess the relationships between environmental factors by the different distances, principal component analysis (PCA) was conducted using R software version 4.3.2 with the packages of factoextra, FactoMineR, and ggplots 2. Environmental data in PCA was transformed "z-score." Additionally, redundancy analysis (RDA) was performed using R software version 4.3.2, incorporating the dplyr, reshape2, readxl, ggpubr, ggplot2, vegan, and ggrepel packages to evaluate the influence of environmental factors on phytoplankton genera and FGs. Transformation "z-score" is for environmental data and "hellinger" for phytoplankton data in RDA.

We used the Shannon–Wiener index (Shannon & Weaver, 1949) based on the genera catalogue to calculate the phytoplankton genera diversity in the study area. The diversity index through the Shannon–Wiener index can be considered as a proxy of an ecological diversity index. A one-way ANOVA with Turkey's HSD test was applied to assess the differences in this index following spatial (different distances) and temporal (March and July).

Furthermore, to assess whether the distribution of phytoplankton genera in aquaculture ponds differs from lagoon areas, we utilized the WinTWINS software version 3.0 (Hill & Šmilauer, 2005). WinTWINS originated from TWINSPAN software written in 1979 by Hill et al. and is used for indicator species/genus analysis. The input is relative abundance data on phytoplankton genera formatted in the Fortran language. The choice of analyzing indicator genera was made only in July because at that time, phytoplankton experienced a substantial growth and a pronounced increase in biomass when compared to the period in March. We defined cut levels at 0, 5, 10, 20, and 50, with abundance percentages above 50% assigned a value of 5, while percentages below 50% were classified as 1 (0-4%), 2 (5-9%), 3 (10-19%), and 4 (20-49%).

# Results

Spatial and temporal patterns of the nutrients and environmental factors

The environmental factors showed significant variations and substantial differences between different distances in March and July (Fig. 2).

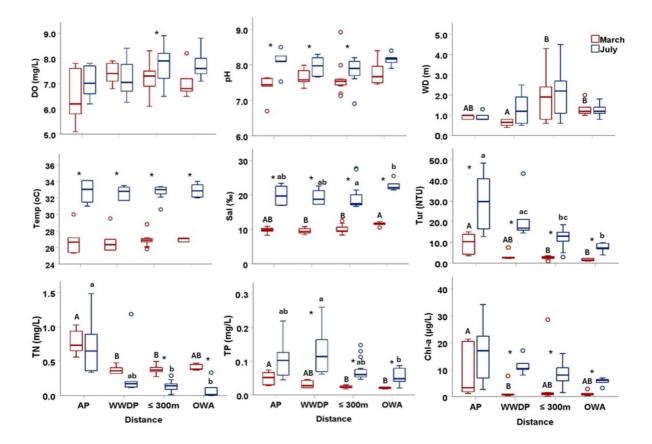


Fig. 2 Box-plots ( $\pm$ SD) showing water quality parameters in aquaculture ponds (AP), wastewater discharge points (WWDP), at a distance of 50–300 m from discharge point ( $\leq$ 300 m), and open water area (OWA) of the Thuy Tu and Cau Hai lagoons in March (in red) and July (in blue) with DO (dissolved oxygen), WD (water depth), Temp (temperature), Sal (salinity), and Tur (turbidity). Red and blue circles were

outliers for environmental parameters in March and in July, respectively. The capitals and lowercases represent a comparison of the environmental parameters at different distances in March and in July, respectively. Asterisk (\*) indicates the statistical difference of environmental parameters in March when compared to July within the same distance class

A Kruskal–Wallis test for environmental factors within the same distance class between March and July revealed that except for water depth, most factors statistically differed between both periods at WWDP,  $\leq$  300 m, and OWA with Asymptotic sig. < 0.04, and even < 0.001 at a distance within  $\leq$  300 m, whereas the aquaculture ponds (AP) differed between both periods for pH (Asymptotic sig. = 0.01), salinity (Asymptotic sig. = 0.004), temperature (Asymptotic sig. = 0.01). DO, pH, salinity, temperature, turbidity, and TP in July were higher than in March, whereas the opposite was true for TN concentration. The comparison of environmental factors between the different distance classes

through the Kruskal–Wallis test showed no apparent differences in DO, pH, and temperature from the aquaculture ponds (AP) to the open water area (OWA) in March and July. However, turbidity in the AP fluctuated within a large range from 3.6 to 15.2 NTU in March and from 12.9 to 48.4 NTU in July, being significantly higher than those within a distance of  $\leq 300$  m (Adj. Sig. of 0.014 and 0.047 for these distances in March and in July, respectively) and at OWA (Adj. Sig. of <0.001 and 0.001, respectively). Additionally, the values of salinity at OWA (mean of  $11.5 \pm 0.5\%$ ) were higher than at WWDP (mean of  $9.6 \pm 0.8\%$ ) with Adj. Sig. of 0.010 and at  $\leq 300$  m (the mean of  $10.1 \pm 1.1\%$ ) with Adj. Sig. of 0.031 in March, whereas these salinity values at OWA were higher than those within a distance of  $\leq 300$  m in July (Adj. Sig = 0.044). TN concentrations in the AP ranged from 0.56 to 1.0 mg/L (mean of  $0.77 \pm 0.18$  mg/L), being sharply higher than those at WWDP (Adj. Sig = 0.005) and within a distance of  $\leq 300$  m in March (Adj. Sig = 0.001), whereas TN concentrations in AP (mean of  $0.73 \pm 0.45$  mg/L) were significantly higher than those at a distance at  $\leq 300$  m and at OWA in July (Adj. Sig of 0.004 and 0.001, respectively). Regarding TP concentrations, these values in the AP ranged from 0.027 to 0.073 mg/L (mean of  $0.050 \pm 0.020$  mg/L), being higher than those in the samples at a distance of  $\leq 300$  m (mean of  $0.024 \pm 0.004$  mg/L) with Adj. Sig = 0.007 and at OWA (mean of  $0.021 \pm 0.003$  mg/L) with Adj. Sig = 0.004 in March. However, TP concentration was significantly different in July between WWDP (the mean of  $0.130 \pm 0.075$  mg/L) and OWA (mean of  $0.054 \pm 0.025$  mg/L) with Adj. Sig = 0.049. The values of Chl-a in the ponds ranged from 1.17 to 21.36 mg/L, which were sharply higher than those at WWDP in March (from 0.39 to 7.69 mg/L) with Adj. Sig = 0.038.

Concerning the relationship between environmental factors by the distinguished distance classes from AP to OWA, the results of the PCA showed that the first two principal components explained over 53% of the data in March and over 63% of the data in July (Fig. 3). The first axis explained 34.0% and 39.7% of the variation in March and July, respectively. Nevertheless, the first axis was positively associated with the data on nutritional factors of TN and TP and turbidity in March whereas it was negatively associated with TN, TP, and turbidity in July. Besides, there was a strong relationship between these three factors and Chl-a in July. Additionally, the nutrients (TN and TP) and turbidity were characteristic factors for the aquaculture ponds. The second axis explained 19.5% of the variation of environmental factors in March and was positively associated with salinity whereas it explained 23.8% and was closely negatively associated with salinity in July. Additionally, the PCA of March also illustrated a strong relationship of DO with pH, which were primary factors characterizing the OWA, while the area within a distance of < 300 m to the aquaculture ponds was featured by lower water depth. By contrast, in the driest moment of the year (July) with an increase in temperature and salinity, there was a relationship between DO, temperature, and water depth within the area  $\leq 300$  m, while salinity was an ultimately influencing factor at OWA.

#### The distribution of phytoplankton

In total, 35 phytoplankton genera were identified in March and July. Several genera were found only in March, including *Spirogyra* spp., *Anabaena* spp., and *Calothrix* spp. These representatives are often distributed in a freshwater environment and areas

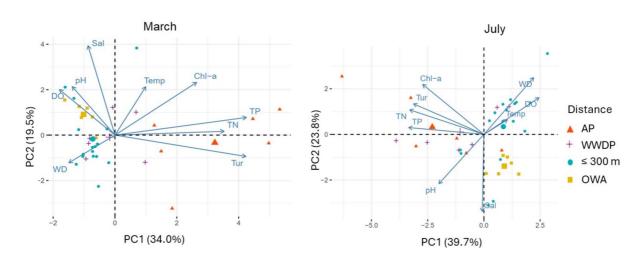


Fig. 3 Biplots of PCA between environmental factors (DO, dissolved oxygen; WD, water depth; Temp, temperature; Sal, salinity; Tur, turbidity) for aquaculture ponds (AP), wastewater

discharge points (WWDP), at a distance of 50–300 m from discharge point ( $\leq$  300 m), and open water area (OWA)

with low salinity. Likewise, several genera were present only in July, including Licmophora, Dictyocha, Akashiwo, and Amphiprora. Most of these are marine phytoplankton distributed in environments with high salinity. A significant variability in mean cell density was observed within the same distance at both moments of the study (Asymptotic sig. < 0.02). It was evident that in March, the mean cell density remained relatively low, staying below 25,000 cells/L. In contrast, in July, the mean cell density was substantially higher, from  $20 \times 10^3$  cells/L to a peak at WWDP of  $139 \times 10^3$  cells/L (Fig. 4). Additionally, we found that in March, the genera of Cylindrotheca and Nitzschia dominated in AP. In contrast, in July, when phytoplankton growth was more developed, the genera of Oscillatoria, Navicula, and Nitzschia were predominant within the ponds. Outside AP, Thalassiosira spp. prevailed, with the highest density at WWDP (ranging from 250 to  $544.10^3$  cells/L) and gradually decreased with distance towards OWA.

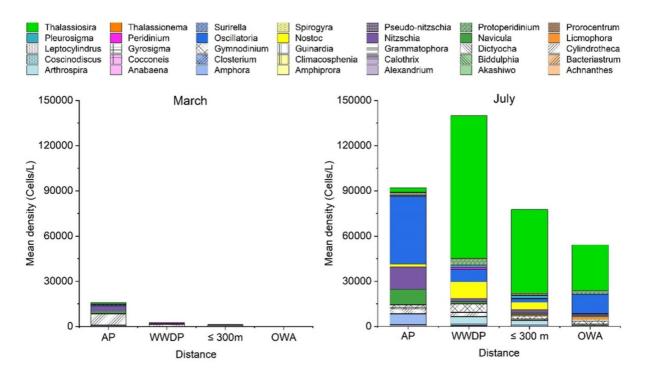
Although the phytoplankton density showed significant differences between March and July and in AP compared to farther distances, the Shannon–Wiener index indicated that differences in phytoplankton genera diversity were only observed at OWA in March compared to July and other distances in March. In contrast, no differences in the diversity of phytoplankton genera were found in July (Table 2).

The results of the RDA, depicting the effect of environmental factors to the composition of phytoplankton genera (Fig. 5), indicated that the first two axes (RDA1 and RDA2) explained this relationship

Table 2 The mean $\pm$ SD of the Shannon–Wiener index by the different distances in March and July

Distance	Shannon-Wiener	index
	March	July
AP	$1.5 \pm 0.4^{b}$	$1.5 \pm 0.5^{b}$
WWDP	$1.6 \pm 0.4^{b}$	$1.5 \pm 0.5^{b}$
≤300 m	$1.3 \pm 0.5^{b}$	$1.6 \pm 0.5^{b}$
OWA	$0.6 \pm 0.5^{a}$	$1.8 \pm 0.6^{b}$

AP, aquaculture ponds; WWDP, wastewater discharge points;  $\leq 300$  m, at a distance of 50–300 m from discharge point; OWA, open water area. The different letters indicate statistical differences between distances among each other and between March and July within the same distance



**Fig. 4** Mean phytoplankton density in March and July for aquaculture ponds (AP), wastewater discharge points (WWDP), at a distance of 50–300 m from discharge point ( $\leq$  300 m), and open water area (OWA)

#### Deringer

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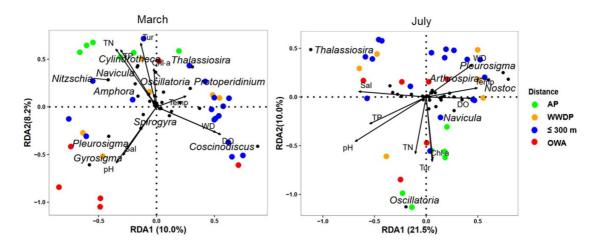
better in July (over 31%) compared to March (18.2%). Similar to the results in Fig. 4, we found that the factors of nutrients (TN, TP) impacted the contribution of *Cylindrotheca* spp., *Navicula* spp., and *Nitzschia* spp. in March, whereas only *Oscillatoria* spp. was influenced by these factors in July. Furthermore, water depth and DO influenced the composition of *Coscinodiscus* spp. in March, whereas the genera of *Nostoc* and *Pleurosigma* were impacted by water depth, temperature, and DO in July.

The habitat preferences and representative genera of dominant FGs observed in this study are presented in Supplementary material. When considering the dominance of FGs by the different distance classes in March and July (Fig. 6), we found that the MP group was the dominant group in both March and July, especially in AP, accounting for 88.2% and 85.4%, respectively. Notably, in July, the MP group was the most prevalent across all distances, accounting for 85.4 to 88.9%. The H2 group was primarily distributed at WWDP and at  $\leq$  300 m (7.9% and 6.2%, respectively), while at the OWA locations, the dinoflagellate groups were prevalent (5.0%). Furthermore, in July, we found that there was an appearance of phytoplankton in the two groups SMD and MS, which were absent in March. SMD and CPMD groups had the highest density at OWA (accounting for 3.0% and 1.5%, respectively) compared to their distribution at other distances. The RDA results in March and July (Fig. 7) revealed that two axes RDA1

and 2 in July explained 38.4% of the relationship between FGs and environmental factors by the different distances, more substantial than the explained variation in March (27.5%). In both March and July, we found that MP was influenced by the nutrients (TN and TP).

#### Indicator genus analysis

According to the results of the indicator genus analysis (Fig. 8 and Supplementary material), we found that Division 1 effectively segregated the samples into two spatial distinct groups. The arbitrarily defined negative group comprised 20 samples, including all AP and the sampling points in Thuy Tu lagoon. On the other hand, the positive group consisted of 15 samples from the Cau Hai lagoon and the locations at the connecting area between Thuy Tu and Cau Hai lagoons (collectively referred to as the Cau Hai lagoon). Furthermore, by analyzing the results of Divisions 1, 2, and 4, we identified that Pleurosigma and Nostoc served as indicator genera for Thuy Tu lagoon. Regarding the Cau Hai lagoon, there were many indicator genera in the result of Division 1 (Thalassiosira, Licmophora, Akashiwo, Gymnodinium, and Protoperidinium). However, according to Division 3, although no indicator genera in nine sites in Cau Hai lagoon, Gymnodinium spp. and Nitzschia spp. were indicators for the remaining six sites. Therefore, we suggest that Gymnodinium spp.



**Fig. 5** Biplots of RDA between environmental factors and phytoplankton in March and July for aquaculture ponds (AP), wastewater discharge points (WWDP), at a distance of

50–300 m from discharge point ( $\leq$  300 m), and open water area (OWA) with DO (dissolved oxygen), WD (water depth), Temp (temperature), Sal (salinity), and Tur (turbidity)

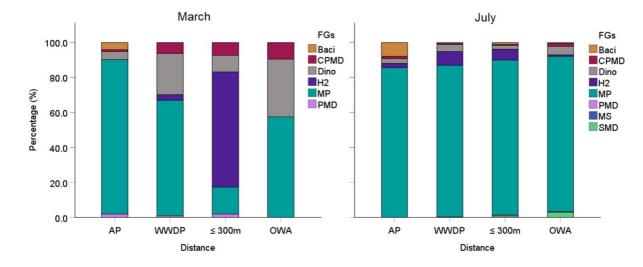


Fig. 6 Distribution of functional groups (FGs) in March and July for aquaculture ponds (AP), wastewater discharge points (WWDP), at a distance of 50–300 m from discharge point ( $\leq$  300 m), and open water area (OWA). Functional groups are MP (inorganically turbid shallow lakes), H2 (mesotrophic

shallow lakes), CPMD (centric planktonic marine diatoms), PMD (pennate marine diatoms), Dino (dinoflagellates), Baci (Bacillariophycidae), MS (marine silicoflagellates), and SMD (stalked marine diatoms). Details of functional groups and included genera are provided in Supplementary material

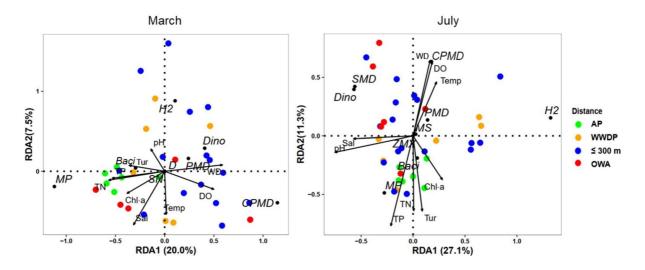


Fig. 7 Biplots of RDA between environmental factors and functional groups in March and July for aquaculture ponds (AP), wastewater discharge points (WWDP), at a distance of 50–300 m from discharge point ( $\leq$  300 m), and open water area (OWA) with DO (dissolved oxygen), WD (water depth), Temp (temperature), Sal (salinity), and Tur (turbidity). Functional

groups are MP (inorganically turbid shallow lakes), H2 (mesotrophic shallow lakes), CPMD (centric planktonic marine diatoms), PMD (pennate marine diatoms), Dino (dinoflagellates), Baci (Bacillariophycidae), MS (marine silicoflagellates), and SMD (stalked marine diatoms). Details of functional groups and included genera are provided in Supplementary material

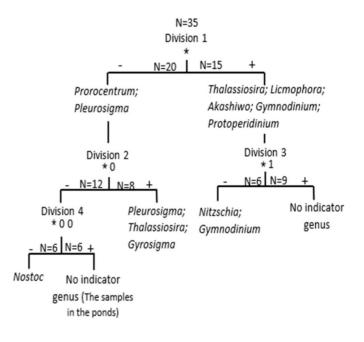
can be indicative in the Cau Hai lagoon. Additionally, according to the Division 4 results, we found that no genera were serving as indicators for the aquaculture ponds.

#### Discussion

A decrease in TN, TP, and Chl-a concentrations was revealed from the ponds towards farther distances.

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Fig. 8 Dendrogram of indicator genera according to a two-way indicator species analysis (TWINSPAN). A detailed matrix of samples and genera is provided in the Supplementary material



These are related to the nutrients of the aquaculture ponds and originated from residual food and fish/ shrimp waste (Erondu & Anyanwu, 2005; Jegatheesan et al., 2011; Kawasaki et al., 2016). Additionally, fertilizers are frequently introduced into aquaculture ponds to support algae growth (Boyd, 2018; Green, 2022). According to disclosures of the Thua Thien Hue Sub-department of Fisheries, during aquaculture, the fertilizers were introduced into the ponds to create a favorable environment for the growth of phytoplankton. The various sources of nutrients made a significant contribution into the nitrogen and phosphorus concentrations in the aquaculture ponds.

Additionally, we found a significant difference in concentrations of TN, TP, and Chl-a and phytoplankton cell density outside of the aquaculture ponds between March and July. The characteristics of aquaculture activities in the Tam Giang-Cau Hai lagoon system were the primary reason for this difference. In practice, aquaculture operations in the Tam Giang-Cau Hai lagoon system typically involve two cultivation periods each year, known as primary cultivation and a supplementary one. The primary cultivation commences in March after the aquaculture ponds are cleaned and dredged to remove accumulated waste sediment, followed sun exposure to eliminate bacteria. Once this preparation is complete, the ponds are filled water, and the fingerlings are introduced. In contrast, the supplementary cultivation starts around July, when aquaculture ponds do not require extensive cleaning, and fingerlings are introduced directly into the ponds. In the supplementary cultivation, the ponds are maintained with lower stocking densities compared to the primary cultivation, and hence, the feeding regime is also less intensive. This likely explains why the TP concentration in July was higher compared to March. It is also associated with a bulk of phytoplankton density in July compared to March. Regarding the lower TN concentration in July compared to that observed in March, the assimilation of nitrogen (nitrate and ammonia) by several phytoplankton species (Caperon, 1968; Eppley et al., 1969; Fernandes et al., 2019; Stenow et al., 2023) and nanopico plankton (Panthalil et al., 2023) could potentially explain this observation.

The nutrients and environmental factors significantly impact the distribution and density of phytoplankton genera. Previous research also demonstrated that nutrients (phosphate and nitrogen) and other environmental factors such as temperature, salinity, total suspended solids (TSS), pH, DO, water depth, and turbidity play an essential role in the structure of phytoplankton communities (Arumugam et al., 2016; Chibsa et al., 2023; Kahsay et al., 2022; Li et al., 2023; Nassar et al., 2015; Sathish Kumar et al., 2023; Sun et al., 2022). The study of Sathish Kumar et al. (2023) on the Southeast coast of India showed that phytoplankton density ranged from 0.7 to  $210 \times 10^3$  cells. $L^{-1}$  and had higher abundance in the nearshore than the offshore stations due to the nutrient input from the land. In our study, the dilution of nutrient concentrations mentioned above could explain the difference and lowering in the phytoplankton cell density with increasing distance towards the open water area. Another study in the Northern East China Sea found that cyanobacteria habitats were characterized by high temperature, high salinity, low concentration of nutrients, and depth, whereas the distribution of diatoms was associated with low temperatures and high nutrient concentrations (Kim et al., 2020). This result was similar to our study, which mentioned environmental factors affecting the distribution of Nostoc (cyanobacteria) in July. Still, it distinguished from the distribution of several diatoms encompassing Coscinodiscus (in March) and Pleurosigma (in July).

The tidal movements and increasing salinity also should be regarded as an important factor impacting the distribution of marine phytoplankton in our study area. Although both Thuy Tu and Cau Hai lagoons are influenced by the tide, the Cau Hai lagoon experiences stronger tides due to its proximity to the Tu Hien inlet. Therefore, the salinity in the Cau Hai lagoon, especially at OWA points, is often higher than in the Thuy Tu lagoon area, hence its influence on the dominance of marine phytoplankton. The selection of taking the samples in March and July is justified by considering the significant environmental differences between these two periods. In March, the region experiences the transition from the wet season to the dry season, whereas July marks the peak of the dry season with the highest temperature and salinity (Tran et al., 2010). These seasonal shifts result in distinct development of the phytoplankton composition, with the appearance of certain genera that thrive in brackish conditions in July (such as Licmophora, Dictyocha, and Akashiwo), whereas March witnesses the presence of genera more suited to freshwater environments with lower salinity (Calothrix, Anabaena, and Spirogyra) due to the river water input. The research of Draredja et al. (2019) showed a seasonal fluctuation in phytoplankton taxa in Mellah lagoon, Algeria. The diatoms were predominant in spring, whereas dinoflagellates developed dominantly in summer and early autumn. These results could be related to the fluctuation of salinity and temperature (Draredja et al., 2019). Trombetta et al. (2019) suggested that the growth and spatial distribution of phytoplankton in coastal aquatic ecosystems are subjected to notable influences from ambient temperature, primarily due to its impact on the photosynthetic processes of these primary producers. Concerning salinity, each phytoplankton species exhibits distinct responses to variations in salinity (Larson & Belovsky, 2013). This led to salinity impacting the structure and composition of phytoplankton communities. The contribution of marine phytoplankton in July also significantly increased the diversity index at OWA in July compared to March. Moreover, the seasonal transition from the rainy to the dry season mentioned above resulted in a lower diversity of phytoplankton genera at OWA in March compared to other distances. Therefore, the development and distribution of phytoplankton are closely linked to both nutrients and environmental conditions.

The distribution of environmental factors and nutrient conditions by the various distances and during the two sampling periods has led to a variation among the FGs. The primary criterion for categorizing phytoplankton into functional groups relies on the similarity among phytoplankton, including their morphological features and habitat adaptation traits. Besides other ecological features (e.g., phenology), species with similar seasonality may respond similarly to specific environmental conditions (Salmaso et al., 2015). This approach not only streamlines the examination of phytoplankton communities but also effectively elucidates the correlation between phytoplankton and environmental factors (Becker et al., 2009; Kruk et al., 2017). In our study, the genera of the MP group are predominantly distributed in nutrient-rich areas, often within the aquaculture ponds. Members of the MP group are also observed at other distances and exhibit a dominant advantage over other FGs. This suggests the possibility of frequent water exchange between the aquaculture ponds and the lagoon water. Our study showed that FGs representing marine phytoplankton (CPMD and SMD) had their highest density at OWA compared to other locations as well as the appearance of MS and SMD just in July highlights the influence of salinity on the development of marine phytoplankton. The OWA region in the Cau Hai lagoon, influenced by tides and the interaction with the sea through the Tu Hien inlet, must have played a significant role in this distribution pattern.

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Pleurosigma spp. and Nostoc spp. served as more suitable indicators for water quality in the Thuy Tu lagoon, whereas no indicator genera or species were found for the ponds. The results of the indicator genus analysis for July are in complete accordance with the RDA results of phytoplankton genera. The spatial and temporal variation may lead to the difference in dominant genera that can become indicators in the Thuy Tu and Cau Hai lagoons. Thalassiosira is a genus with a vast distribution range. Thalassiosira spp. appear in many estuaries, coasts, and oceans (Park et al., 2016). Besides, Fernandes et al. (2019) suggested that Thalassiosira spp. were abundant in white-leg shrimp ponds. In our study, this genus also served as an indicator for the sites in the Cau Hai lagoon (the positive group at division 1), as for eight sites in the Thuy Tu lagoon (the positive group of division 2). The eight sites in Thuy Tu lagoon mentioned above comprised one sample at a WWDP and seven samples at  $\leq 300$  m. With regard to *Pleu*rosigma and Nostoc, these two genera exhibited high levels of abundance at WWDP and  $\leq$  300 m locations in the Thuy Tu lagoon. The RDA results of July confirmed this observation. Pleurosigma spp. (e.g. P. salinarum and P. elongatum) were suggested to have high tolerance ranges of physio-chemical variation in water conditions (Dalu et al., 2016). Besides, Pleurosigma spp. could be dominant in areas with high nutrient concentrations, such as aquaculture ponds (Fernandes et al., 2019; Supono & Hudaidah, 2018). Nostoc is a common genus in terrestrial and aquatic habitats (Dodds et al., 1995; Potts, 2002). Mollenhauer et al. (1999) found that there were two species distributed in highly eutrophic, polytrophic, or even saprobic systems due to anthropogenic pollution (Nostoc caeruleum) and experimentally fertilizing (Nostoc pruniforme). Six sites featuring Nostoc spp. as an indicator were located at all wastewater discharge points in Thuy Tu lagoon. In accordance with Mollenhauer et al. (1999), this may support our interpretation of the distribution of Nostoc spp. around the areas influenced by aquaculture pond fertilization. Our study revealed that the phytoplankton assemblages in the ponds are not unique but contain genera that are also present in other parts of the lagoon. This improved our view that there is effective mixing and regular water exchange between aquaculture ponds and water outside them in Thuy Tu and Cau Hai lagoons, which diminishes the specificity of the pond conditions. This water exchange also confirms the impact of aquaculture wastewater on the lagoon's water quality. It is the basis for assessing ecological risks caused by aquaculture and proposing appropriate solutions.

Phytoplankton serves as an environmental indicator in developing aquaculture and assessing the environment. Although phytoplankton contributes to primary productivity that plays an ultimate role in sustaining the fishing industry (Brraich & Saini, 2015) in aquaculture ponds, several species belonging to genera such as Gymnodinium, Alexandrium, and Pseudo-nitzschia are harmful species and can pose threats to seafood species in the ponds (Dorantes-Aranda, 2023; Dorantes-Aranda et al., 2015; Hernández-Sandoval et al., 2022). Therefore, phytoplankton monitoring contributes to an increased awareness of local stakeholders about potential toxic species associated with aquaculture ponds. Besides, phytoplankton data can support the assessment of the nutrient status in the lagoon and propose effective environmental monitoring strategies. In particular, phytoplankton monitoring indicated a substantial marine influence in the more open waters, thereby highlighting the importance of maintaining an inlet at the ocean and avoiding closure through sedimentation of the bordering dune landscape.

# Conclusion

Understanding how environmental factors affect the distribution and density of phytoplankton communities has shed light on the impacts of aquaculture on the study area. The elevated phytoplankton density is evident in aquaculture ponds and their discharge channels, even though precise indicator species for aquaculture ponds remained unidentified. We found that, in addition to the influence of nutrients originating from aquaculture activities, external environmental factors such as seawater also significantly influence phytoplankton distribution. Therefore, the characteristics of the phytoplankton community could serve as valuable evidence for local stakeholders to comprehensively monitor aquaculture ponds and assess the environmental quality of the Tam Giang-Cau Hai lagoon system. Besides, the role of marine phytoplankton in the lagoon system's food chain should be thoroughly considered and assessed to support effective policy in environmental management.

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**Data availability** All data supporting the findings of this study in its current form are available within the paper and supplied supplementary materials.

#### Declarations

**Ethics approval** Not applicable: our manuscript does not report on or involve the use of any animal or human data or tissue.

**Conflict of interest** The authors declare no competing interests.

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Page 17 of 17 **1099** 

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