

The Seasonal Changes of Phytoplankton Community in Dongzhai Harbor Mangrove Reserve, Hainan, China

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Abstract: An essential function of phytoplankton is to facilitate the movement of materials, energy, and information within mangrove ecosystems. Because they are sensitive to changes in the ambient conditions of aquatic bodies, phytoplankton is frequently employed for environmental monitoring in aquatic ecology. By sampling at six stations in October (the normal season) and December (the dry season) in 2020, the spatial and temporal variations of phytoplankton as well as the effects of environmental variables on phytoplankton community structure were evaluated in order to better understand the dynamics of the phytoplankton community in Dongzhai Harbor Mangrove Reserve, Haikou, China. The findings demonstrated that phytoplankton's species richness, cell density, and diversity revealed temporal and spatial change. During the normal season, green and blue-green algae predominated, but during the dry season, cryptophytes replaced them. The phytoplankton community's seasonal average dissimilarity was 58.23. The Dongzhai Harbor Mangrove Reserve's phytoplankton community was found to be impacted by water temperature, turbidity, and total nitrogen, according to RDA results. This is the first study simultaneously investigating the phytoplankton community and their environment in this area, and it is essential in order to set up the baseline for future studies.

Keywords: Phytoplankton; Environmental factors; Dominant species; Water quality assessment

1. Introduction

Mangroves are composed of including trees, shrubs, palms, and ferns that growing in the inter-tidal zone of tropical and subtropical coasts, which suffer from periodic tide^[1]. Mangrove ecosystem plays multiple ecological functions such as material production, windbreak and sand fixation, water purification^[2-3]. The ecosystem harbors rich diversity because of dynamic heterogeneity^[3-4], Kathiresan^[4] found 13 species of mangrove trees and 819 other species at a tropical mangrove ecosystem. He et al^[5] reported that mangrove wetlands in China is 1766 times of species abundance per unit area than ocean. However, over the last two decades, mangroves are considered to be highly degraded coastal ecosystems because of anthropogenic activities such as aquaculture, farming, the process of urbanization, exploitation of tourism, over-harvesting and coastal pollution^[6]. The phytoplankton usually are used as the ecological indicator to decipher because phytoplankton species composition have the rapid response to the change of the ecological conditions and trophic status of the water body. Some studies about phytoplankton community have showed mangrove ecosystem harbor a wide variety of phytoplankton species. Gao et al^[7] indicated that *Conticribra weissflogii* and *Cyclotella atomus* are pollution indicative species of sewage flow in Futian Mangrove of Shenzhen, China. Pham^[8] found phytoplankton assemblage in the Can Gio Mangrove Biosphere, Vitenam was influenced by salinity, nitrate and phosphate concentration. Inyang & Wang^[9] revealed the impact of reactive nitrate, phosphate, and turbidity on the phytoplankton community structure in mangrove zones of Guangzhou Province, China. Some research on the estuary and coastal waters demonstrated that water temperature^[10], dissolved oxygen and pH were responsible for the variations of phytoplankton community structure^[11]. Given these varied results, we need further efforts to explore the key environmental factors driving phytoplankton community in mangrove ecosystems.

Hainan Island, located in the southernmost region of China, has a tropical marine monsoon climate and is known for its rich biodiversity and mangrove forests^[12]. The Dongzhai Harbor Mangrove Reserve (DHMR, 110°32'E~110°55'E, 19°51'N~20°01'N), established in 1980 in the northeastern part of the island, is the largest and most pristine natural mangrove area in China. Covering a total area of 3337.6 hm², of which about 47% is mangrove, it is not only the first mangrove nature reserve in the country, but also one of the seven protected areas on the International List of Important Wetlands. DHMR is home to the largest and most complete collection of mangrove plants in China, with 35 species belonging to 19 families. Despite extensive research on the reserve's biodiversity, including meiofauna and bottom ciliates, the phytoplankton population remains understudied and lacks sufficient scientific data. Unfortunately, the mangrove forests lining the estuarine beaches have been increasingly threatened in recent years by the rapid expansion of aquaculture, tourism and urban development along Hainan's coastal regions. Despite these challenges, the reserve remains a vital habitat for diverse and endemic species, contributing significantly to China's ecological balance and biodiversity. Our research attempts to: 1) investigate the phytoplankton community structure in Dongzhai Harbor Mangrove; and 2) show how the environmental elements in an estuarine-mangrove ecosystem relate to the phytoplankton community; 3) explore the effects of seasonal changes on phytoplankton communities.

2. Materials and methods

2.1. Sample collection and laboratory experiment

We have selected six stations in DHMR in Hainan Island (110°32'E~110°55'E, 19°51'N~20°01'N) (Fig. 1) were Tashi Station (TS), Sewage Plant (WS), Mangrove Protection Bureau (BH), Shanwei Village (SS), Daoxue Station (DX) and Sanjiang Gate (SJ). In 2020, the samplings were conducted in October and December. HACH SL1000 was used to measure the physicochemical properties of the water, such as pH, conductivity, dissolved oxygen, and water temperature. The Secchi disc was used to measure Secchi depth (SD), the ATAGO POCKET SALT METER was used to test salinity (Sal), and the HACH 2100Q Portable Turbidity-meter was used to measure turbidity (Tur). Total phosphorus (TP) and total nitrogen (TN) were measured and tallied in accordance with Chinese inquiry guidelines. At six stations, we used containers to gather 1 L of phytoplankton samples from the water at a depth of 0.5 m below the surface. Following a 48-hour sedimentation period, we concentrated the samples to 50 milliliters and conserved them by adding 1% Lugol's iodine solution all at once. Under Nikon ECLIPSE E100 with 400x magnification and consistent mixing, the condensed sample was counted in a 0.1 ml counting chamber. Phytoplankton's identification was according to the standard works by Hu & Wei^[13] and Guo^[14].

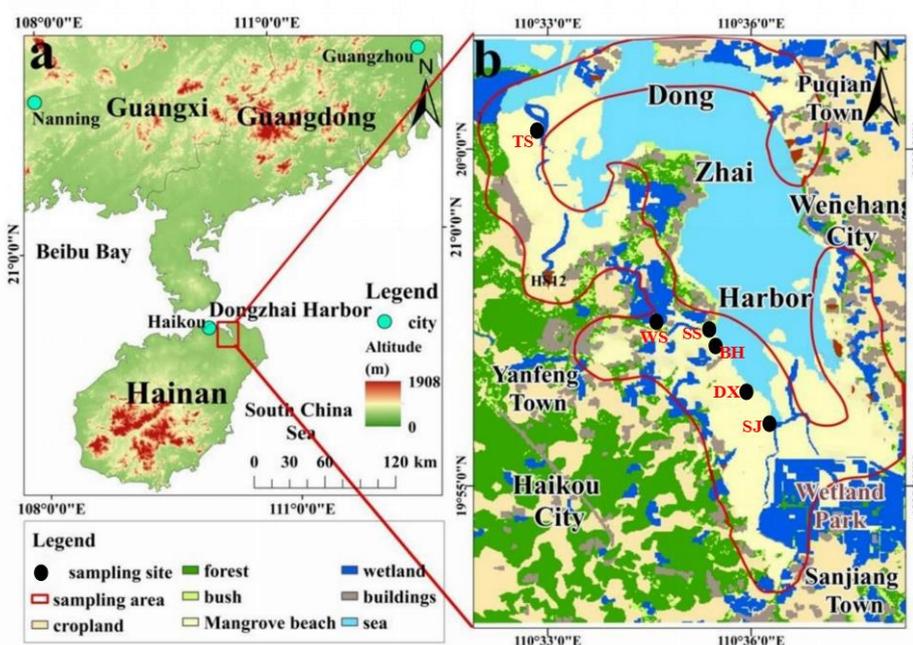


Figure 1: The sampling sites of phytoplankton in DHMR during the 2020(referenced from Zhang et al., 2022^[15]).

2.2. Statistical analysis

The physico-chemical and biological parameters in the samples in DHMR were correlated with environmental variables, phytoplankton abundance, and environmental factors using a Pearson correlation analysis. The analysis of similarity (ANOSIM) test was employed to evaluate the degree of community structure similarity among the samples, after the non-metric multidimensional scaling (NMDS) test. Based on the breakdown of the Bray-Curtis dissimilarity index, several significant species were found using the similarity percentages analysis (SIMPER)^[16]. The phytoplankton species and environmental conditions were subjected to the redundancy analysis (RDA). R software was used for all of the aforementioned procedures^[17].

3. Results

3.1. Community composition of phytoplankton

During the investigation, a total of 168 species were reported comprising 81 species of Bacillariophyta, 59 species of Chlorophyta, 19 species of Cyanophyta, 4 species of Euglenophyta, 2 species of Dinophyta, 2 species of Cryptophyta, only 1 species of Chrysophyta. *Cyclotella meneghiniana*, *Synedra acus*, *Navicula capitata*, *Cocconeis placentula*, *Chlorella vulgaris* phytoplankton species were found in all stations of study area.

The spatiotemporal fluctuations in phytoplankton abundance in DHMR. The phytoplankton abundance varied between 1.48×10^5 and 3.05×10^6 cells/L, peaking in October at TS station and falling off in WS station in December. The three main groups in October were Bacillariophyta, Cyanophyta, and Chlorophyta, with respective percentages of 47%, 30%, and 20%. With 44% and 37% of the total, Bacillariophyta and Cryptophyta were the two most prevalent groups in December. During the dry season, cryptophytes took the place of blue-green algae and green algae, which were dominant during the regular season. According to the spatial variation, Cyanophyta made up a larger contribution at BH, DX, and SJ, while Bacillariophyta dominated at TS, WS, and SS station.

The dominant species of phytoplankton existed temporal and spatial change (Fig. 2). The major discriminating species found in October were cyanophytes such as *Merismopedia tenuissima*, *Pseudanabaena*, *Oscillatoria princeps* and diatoms such as *Coscinodiscus wailesii*, *Skeletonema costatum*, however the cryptophytes and pennatae diatoms occupied dominance in December.

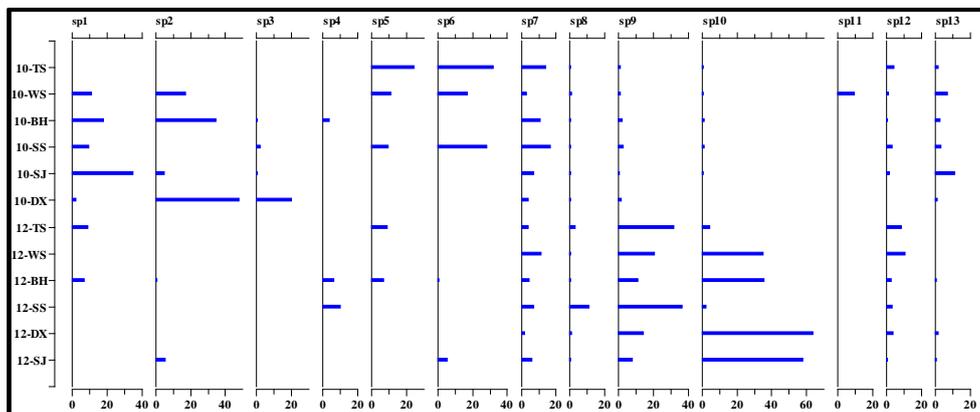


Figure 2: The dominant species of phytoplankton in DHMR

The spatiotemporal patterns of phytoplankton alpha diversity emerged (Fig. 3). Throughout the study duration, no significant variations were observed in the Pielou index ($p > 0.05$), whereas the Shannon-Wiener diversity index exhibited statistically significant differences ($p < 0.05$). In detail, the Shannon-Wiener diversity index (H') of phytoplankton fluctuated between 1.33 and 2.09. In October, the peak diversity index (H') of 2.09 was recorded at SJ, whereas the nadir of 1.77 was observed at DX. Similarly, in December, the highest diversity index of 1.87 was noted at TS, and the lowest value of 1.33 was again recorded at DX. Furthermore, the species evenness (J') of phytoplankton varied considerably, ranging from 0.5 to 0.82. In October, WS exhibited the lowest evenness (J') of 0.5, whereas SJ demonstrated the highest evenness of 0.72. Conversely, in December, WS showed the highest evenness of 0.82, and DX exhibited the lowest evenness of 0.55. A comprehensive overview of these calculated diversity indices is

presented in Table 1.

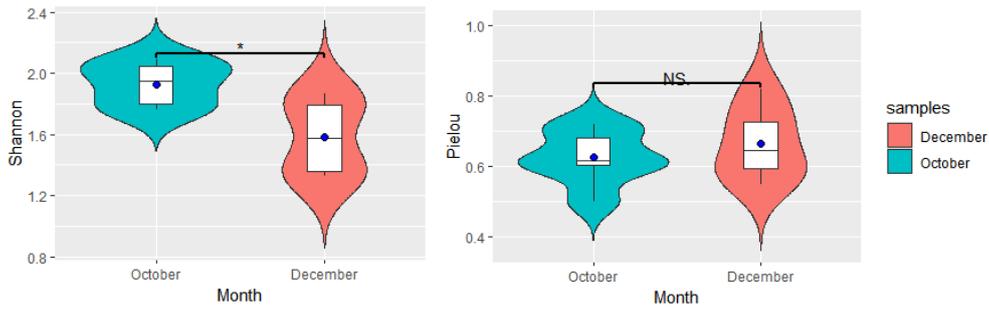


Figure 3: The alpha diversity index of phytoplankton in DHMR.

Table 1: The average temporal variation of alpha diversity indices (H' , J') in the DHMR.

		H'	J'
October	TS	2.06	0.62
	WS	1.90	0.50
	BH	1.77	0.60
	SS	2.00	0.70
	DX	1.76	0.61
	SJ	2.09	0.72
December	TS	1.87	0.74
	WS	1.34	0.82
	BH	1.75	0.69
	SS	1.81	0.6
	DX	1.33	0.55
T Test	t	-2.93	0.75
	p	0.02	0.47

3.2. Relationship between Phytoplankton and Environmental Factors

The phytoplankton cell density and environmental parameters were correlated using a correlation analysis (Fig. 4). The findings indicated that there was a substantial positive association ($p < 0.05$) between the cell abundance of Cyanophyta and WT, SD, and TN. The cell abundance of Bacillariophyta exhibited a substantial negative correlation ($p < 0.05$) with TP, while the cell abundance of Chlorophyta exhibited a positive association ($p < 0.05$) with WT.

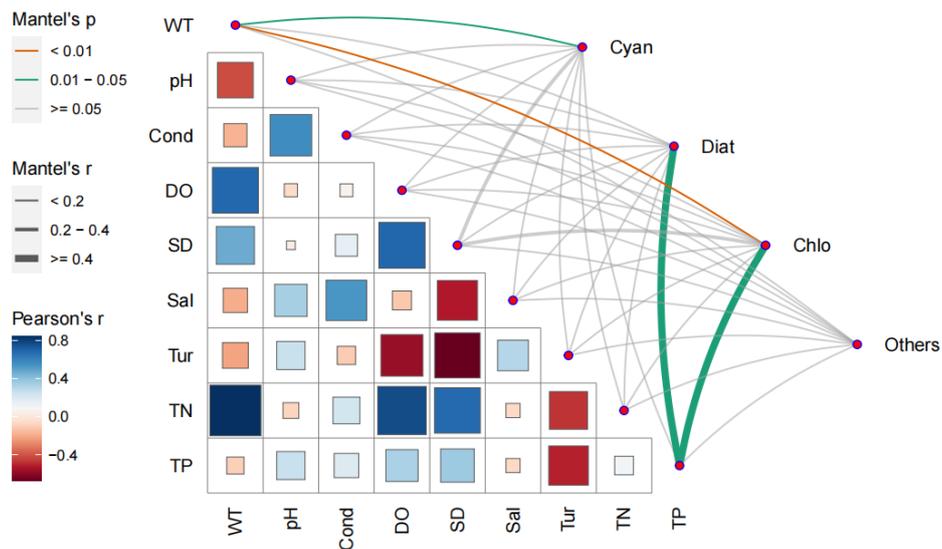


Figure 4: The correlation between phytoplankton abundance and environmental factors

Redundancy analysis (RDA) was used to examine the relationship between dominant phytoplankton species and environmental variables, and the longest axis length was found to be 3.26 based on the results of detrended correspondence analysis (DCA) (Fig. 5). Axis 1 and Axis 2 had an explanation of 31.56% and 22.51%, respectively. Utilizing regression analysis, it was demonstrated that the WT ($r^2=0.87$, $P<0.01$), DO ($r^2=0.56$, $P<0.05$), SD ($r^2=0.54$, $P<0.05$), and TN ($r^2=0.69$, $P<0.01$) were clear explanatory environmental variables that explained phytoplankton distribution and abundance.

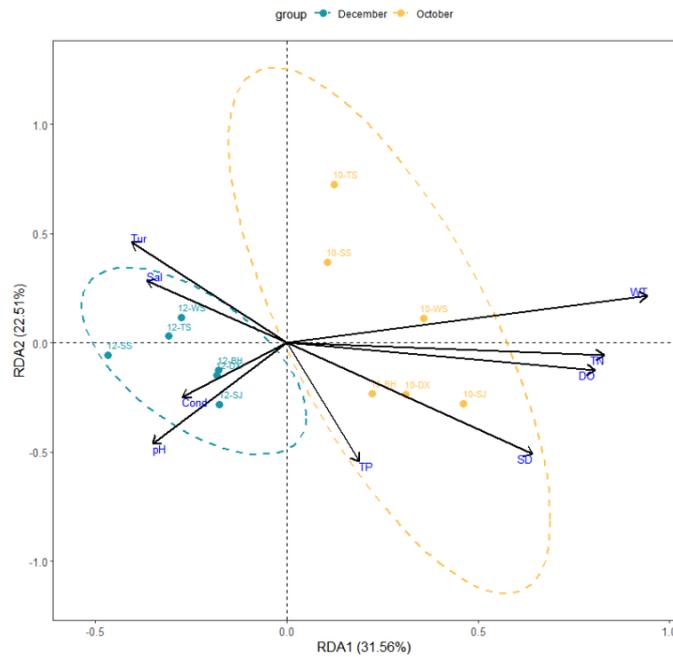


Figure 5: The ordination of Redundancy analysis (RDA) between environmental factors and phytoplankton dominant species.

3.3. The Effects of Seasonal Changes on Phytoplankton Communities

The water temperature (WT), dissolved oxygen (DO), total nitrogen (TN) were significantly higher in October than in December (Table 2). The WT, DO and TN value were higher in October compared to in December. WT at the study locations ranged from 22.2°C to 28.9°C with an average of 25.64°C. DO at the study locations ranged from 6.52 mg/L to 8.56 mg/L with an average of 7.65 mg/L. TN at the study locations ranged from 2.69 mg/L to 3.33 mg/L with an average of 3.09 mg/L.

Table 2: Average, standard error values of different environmental and *t* test of DHMR in October and December in 2020.

	October	December	T test	
			t	p
WT	28.58±0.32	22.7±0.5	-24.37	<0.001
pH	7.78±0.09	7.86±0.12	1.24	0.25
Cond	25.25±5.17	26.98±7.34	0.47	0.65
DO	8.1±0.34	7.19±0.67	-2.94	<0.05
SD	35.83±3.76	33±3.16	-1.41	0.19
Sal	1.96±0.62	2.06±0.67	0.27	0.79
Tur	17.15±6.89	19.12±7.61	0.47	0.65
TN	3.25±0.1	2.93±0.12	-4.91	<0.001
TP	0.39±0.22	0.42±0.18	0.35	0.81

Based on the relative quantity of phytoplankton reported by the discriminating species, the SIMPER procedure was examined (Table 3). The seasonal average dissimilarity was 58.23. *Chroomonas acuta* (0.16, 20.5%), *Navicula* sp. (0.09, 11.9%), *Pseudanabaena* sp., *Skeletonema costatum*, *Merismopedia tenuissima*, *Coscinodiscus wailesii*, *Cyclotella meneghiniana*, *Scenedesmus quadricauda*, *Aphanizomenon* sp., *Chlorella vulgaris* were the major contributors to the seasonal changes. The major discriminating species found in October were cyanophytes such as *Merismopedia tenuissima*, *Pseudanabaena*, however the cryptophytes and pennatae diatoms occupied dominance in December.

Table 3: The SIMPER analysis depicted the 'discriminating species' that contribute to the maximum dissimilarity between the seasons

Dissimilarity	Discriminating Species (Average Dissimilarity and Contribution Percentage)
October vs. December (58.23%)	<i>Chroomonas acuta</i> (0.16, 20.5%) <i>Navicula</i> sp.(0.09, 11.9%) <i>Pseudanabaena</i> sp.(0.09, 11%) <i>Skeletonema costatum</i> (0.07, 12.4%) <i>Merismopedia tenuissima</i> (0.06, 8.6%) <i>Coscinodiscus wailesii</i> (0.04, 4.8%) <i>Cyclotella meneghiniana</i> (0.03, 3.5%) <i>Scenedesmus quadricauda</i> (0.02, 2.8%) <i>Aphanizomenon</i> sp.(0.02, 2.6%) <i>Chlorella vulgaris</i> (0.02, 2.4%)

Non-metric multidimensional scaling (NMDS) illustrates the varied phytoplankton distribution throughout the various study sites (Fig. 6). Based on Bray-Curtis dissimilarity, NMDS was used to quantify the seasonal variations in the phytoplankton community (Figure 6B). In general, the first two NMDS axes (stress=0.12) were used to identify patterns in the composition of phytoplankton communities over the seasons. ANOSIM analysis of the phytoplankton across all research sites showed that no significant difference was found between the sampling sites ($p > 0.1$) with $R = -0.44$, but substantial differences were seen in October and December ($p < 0.001$, Figure 6A) with a high range value of $R = 0.8$.

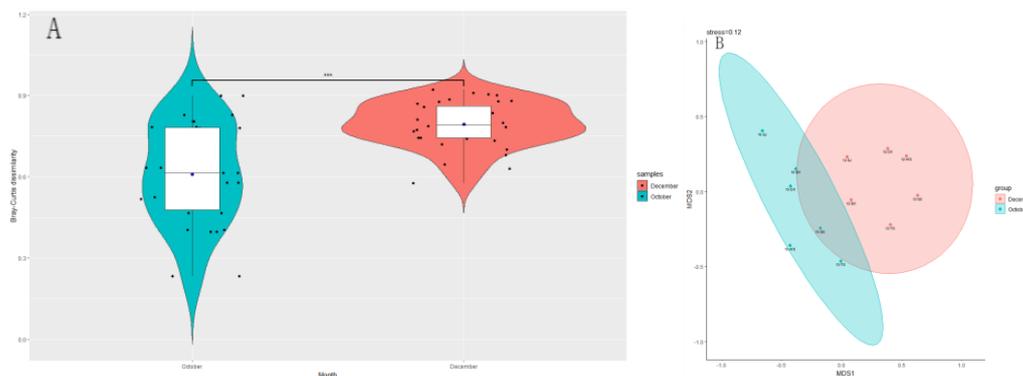


Figure 6: Based on Bray-Curtis dissimilarity in DHMR, seasonal fluctuations and NMDS of phytoplankton beta diversity are presented. The *** ($p < 0.001$) symbol denotes the significance levels.

4. Discussion

4.1. Phytoplankton community composition

The composition of phytoplankton community varies depending on the kind of water^[18-20]. Diatoms make up the majority of the phytoplankton community in the DHMR in this study, which is similar with prior studies from other tropical estuary systems^[21-22]. The *C. wailesii*, *S. costatum*, *N. palea*, *Navicula*, *C. meneghiniana* commonly occurred and became dominant species that reflects the characteristics of the intersection of salt and fresh water in the DHMR region. According to certain study, diatoms may be able to adapt to drastically different hydrographical circumstances, which explains why they are so prevalent in mangrove waters^[23-24]. The majority of common species found in this study were in good agreement with earlier research conducted during this area^[25].

There were notable variations in phytoplankton species between the hydrological seasons. The phytoplankton species richness (168) found in this study was less than that found in the Futian Mangrove in Shenzhen, China^[7, 26], but it was higher than that found in earlier studies (56) in DHMR^[25]. From the normal season to the dry season, the number of species in the Bacillariophyta phylum increased, whereas the Chlorophyta phylum showed the opposite tendency. This may be related to the ecological habits of different algae. Chlorophyta has adapted to exist in warm water, whereas Bacillariophyta were primarily cold-water species^[27-28].

There were differences in the phytoplankton abundance between the normal and dry seasons in DHMR. The results for Shenzhen, China's Futian Mangrove were consistent with the peak abundance

occurring during the regular season^[7]. According to our findings, the dominance of Bacillariophyta, Cyanophyta, and Chlorophyta in the normal season shifted significantly to that of Bacillariophyta and Cyanophyta in the dry season (Figure 4). Changes in water quality and hydrological conditions may be to blame for this shift in the organization of phytoplankton communities. Among all the stations, the maximum cell density was recorded at TS that is closer to marine outer and shrimp culture ponds site, the minimum cell density was recorded at WS that is closer to sewage treatment site. The sampling sites were largely characterized by different dominant species of phytoplankton that indicating different environmental characteristics. Pollution-resistant *C. meneghiniana* and the red-tide species *S. costatum* are the main dominant taxa at TS site in wet season, which indicates that these water bodies may be polluted to a certain extent^[25]. *S. costatum* has been reported to dominate or commonly occur from temperate to tropical estuaries^[21, 29-30] or brackish water. Hilaluddin et al^[31] reported that *S. costatum* could become an indicator for nutrient enrichments. *Pseudoanabaena*, a kind of filamentous cyanobacteria, which is common in reservoirs, fish and shrimp ponds, can be the dominant species at WS, DX sampling sites. Cryptophytes are considered that they are more suitable for water environment with low light and high organic matter concentration. In our study, we found *C. acuta* has the superiority in dry season. Some Chlorophyta species such as *Chlorella*, *Scenedesmus* were also dominant. The entry of fresh water or domestic sewage, which may have resulted from the steady hydrographic conditions at the time, is one reason for this phenomena.

4.2. The relationship between environmental factor and phytoplankton community

Our research results revealed that WT and TN were the main drivers of phytoplankton community expansion during the normal season. Previous research have shown that environmental variables can cause temporal and geographical variation in the phytoplankton community^[32-33]. The predominant phytoplankton species identified in this period were *M. tenuissima* and *Pseudanabaena* sp., due to higher nitrogen concentration and temperature than in dry season. The increasing abundance of cyanophytes and chlorophytes with elevated temperature were implied in various water bodies, including lake^[34], river^[35], tidal mangrove creeks^[36].

While the phytoplankton species composition during the dry season was markedly different from that during the wet season, some characteristic dominating species, such as *C. acuta* and *Navicula* sp., remained the same. According to the RDA results, throughout this time period, the species of the phytoplankton population was favorably correlated with water turbidity. Light might take over as the primary environmental element influencing the dynamics of the phytoplankton community because TN concentrations were higher at this point, which lessened their limiting effect on phytoplankton development. Moreover, *Navicula* prefer environments with inorganically turbid waters that are often agitated up^[37-38]. Because of its ability to adapt to cold climates, *C. acuta* may be able to migrate to an area with better light conditions^[35, 39]. Despite the seasonal variations, diatoms such as *C. wailesii*, *C. meneghiniana* and *Navicula* sp. were always the dominant species, due to their rapid N uptake in nitrate-rich environments and strong intrinsic growth rates^[40-42].

4.3. The effects of seasonal changes on phytoplankton communities

Our research indicates that there are significant differences in plankton abundance and species composition, as well as environmental factors, between the normal and cold seasons. Changes in temperature and precipitation between seasons have led to changes in a number of environmental factors. This affects the composition of the phytoplankton community. We hypothesize that there is an inextricable link with human activity, in addition to the influence of natural environmental factors. Haikou is a famous tourist city in China, and the peak tourist season is from December to February. During this period, the increase in human traffic also brings an increase in environmental pollution and its impact on plankton communities. Numerous studies have shown that human activities are a major threat to biodiversity^[43-45] and that tourism has an impact on the environment^[46]. The results of this study further support our hypothesis that seasonal changes in a range of environmental factors and human activities have a significant impact on phytoplankton community composition.

5. Conclusions

In the present study, phytoplankton had a strong response to seasonal changes, and the phytoplankton population was significantly higher in the normal season than in the dry season. Meanwhile, phytoplankton was strongly influenced by WT, TN and TP, which showed large differences between

seasons.

In summary, this study concluded that seasonal changes have a greater influence on phytoplankton communities.

Acknowledgement

Funding: This project was supported by Hainan Provincial Natural Science Foundation of China (No.322RC739) and the Natural Science Foundation of Qiongtai Normal University (qtnb202206).

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