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# Assessment of threats to a Ramsar site from seafood processing operation effluents

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## Abstract

Waterbodies receiving effluents from seafood processing plants might experience severe eutrophication. The present study was carried out to assess the impacts of effluent discharges on water quality and phytoplankton populations in the Vembanadkol Wetland, located adjacent to seafood processing industries. It was conducted in the Cherthala–Aroor–Edakochi coastal belt of Vembanad Wetland, India, a region containing many processing plants. The trophic status of the lake was assessed with the Carlton trophic state index. Hypereutrophic conditions were observed more in the interconnected channels than in the main waterbody. There was a dominance of Bacillariophyceae, followed by Chlorophyceae, Cyanophyceae and Dinophyceae. Bacillariophyceae, Chlorophyceae and Cyanophyceae were comparatively higher in the interconnected channels, whereas Dinophyceae was high in the main body of the lake. There is a shift in dominance from Chlorophyceae to Cyanophyceae in the interconnected channels, especially in the southern and central portion of the lake. There was a dominance of Cyanophyceae over Chlorophyceae, especially in the interconnected channels, being a characteristic of polluted water. *Nitzschia* sp and *Navicula* sp were abundant in the class Bacillariophyceae. The dominant species in the class Dinophyceae was *Ceratium* sp, which is known to proliferate in nutrient-rich waters. The dominant members of the class Chlorophyceae were *Ankistrodesmus* sp and *Scenedesmus* sp. Furthermore, *Oscillatoria* and *Phormidium* were higher in number in the class Cyanophyceae, indicating the presence of pollutants of biological origin. Waste discharges from the seafood processing industry are a major factor contributing to an alarming rate of organic pollution and subsequent eutrophication in the lake. The results of the present study highlight that further expansion of the seafood industry in the Cherthala–Aroor–Edakochi coastal belt of Vembanad Lake is not desirable.

KEYWORDS
Carlton trophic state index, Ramsar site, seafood processing effluents, Vembanad wetland

## 1 | INTRODUCTION

The seafood industry is considered to exhibit a high employment potential, particularly in rural areas, and also helps preserve foreign exchange reserved, particularly for developing countries. As with most processing industries, seafood processing operations produce a large quantity of waste water containing high contents of organic matter, fat, oil and grease and ammonia–nitrogen (Islam, Khan,

& Tanaka, 2004; Sohsalam, Englande, & Sirianuntapiboon, 2008; Tchoukanova, Gonzalez, & Poirier, 2003). The degree of water contamination depends on the particular processing operation, which might be characterized as of a small magnitude (e.g. washing operation), mild magnitude (e.g. fish filleting) or major magnitude (e.g. blood water drained from fish storage tanks). A related factor aggravating the pollution problem is that receiving waterbodies often do not possess a sufficient water volume to dilute such wastewater inputs to non-polluting levels. Processing seafood products requires large quantities of water, with direct consequences in generating contaminated effluents that can deteriorate the quality of the aquatic environments into which they are discharged (Sirianuntapiboon & Nimnu, 1999). Seafood processing generates potentially large quantities of both liquid effluents and solid wastes containing both organic and inorganic materials from inedible fish parts, and endoskeleton shell parts from the peeling process. In fact, 30%–80% of the raw material in the processing of most fish species for human consumption is waste (AMEC, 2003) consisting of large quantities of organic matter comprising oils, proteins, suspended solids, blood, water, fish shells, small particles of flesh, etc. Large quantities of waste water are also generated through associated activities such as fish unloading, equipment sprays, transportation, cleaning, cooling and production purposes, all generally exhibiting high biological oxygen demands (BOD) and high levels of oil, grease and nitrogen (AMEC, 2003; Champ, O'Connor, & Park, 1981). Effluents from fish and crustacean processing plants are generally characterized by high nutrient concentrations, including high nitrogen contents existing as ammonia (29–35 mg/L), high total suspended solids (0.26–125,000 mg/L), increased BOD (10–110,000 mg/L) and chemical oxygen demand (COD) (496–140,000 mg/L), as well as the presence of sanitizers (AMEC, 2003). Thus, waste waters from seafood processing operations can exhibit high levels of these pollutants (Tay, Show, & Hung, 2006). In fact, data on seafood processing operations indicated a BOD production of 1–72.5 kg of BOD/tonne of product, while whitefish filleting processes typically produce 12.5–37.5 kg of BOD/tonne of product. The BOD occurs primarily as a result of the butchering process and general cleaning, while the nitrogen originates predominantly from blood in the wastewater streams (Environment Canada, 2004). Their high organic matter content frequently contributes to pollution and degradation of the world's oceans and coastlines, particularly near seafood processing units (El-Beltagy, El-Adawy, Rahma, & El-Bedawey, 2005; Ferjani, Ellouze, & Ben Amar, 2005; Moens et al., 2007; Morry, Chadwick, Courtenay, & Mallet, 2003; Sirianuntapiboon & Srikul, 2006; Sohsalam et al., 2008; Tchoukanova et al., 2003).

The negative impacts of seafood processing effluents on water quality were investigated by Walden (1991), Park, Enander, Barnett, and Lee (2001), Morry et al. (2003), Tay et al., 2006, Tchoukanova et al. (2003), Akan, Abdulrahman, Dimari, and Ogugbuaja (2008) and Sankpal and Naikwade (2012). Bonsdorff, Blomqvist, Mattila, and Norkko (1997) reported an increased phytoplankton biomass and decreased species diversity of benthic and fish communities attributable to the discharge of seafood processing wastes to aquatic systems, with seafood-associated pollutants resulting in the destabilization of these waterbodies (DWAF & WRC, 1995; Morrison, Fatoki, & Ekberg, 2001). Excessive abundance of phytoplankton communities can significantly contribute to increased eutrophication of waterbodies (Aktan, Tufekc, Tufekc, & Aykulu, 2005; Ganjian et al., 2009, 2010; Nasrollahzadeh Saravi, Bin Din, Yeok Foong, & Makhlough, 2008).

Kerala is a major sea-front state of India, occupying about nine per cent of the country's coastline and contributing about 22%–31% of coastal fisheries outputs. About one-fifth of its total landmass is comprised of wetlands (Kokkal, Harinarayanan, & Sabu, 2008). There are about 287 seafood exporters in Kerala, with 124 processing plants and 169 cold storages. About 119 pre-processing units are located in the Alappuzha (Alleppey) District, Kerala (Sathyan, Afsal, & Thomas, 2014). Kerala accounted for 10, 8,616 t of seafood products, valued at Rs. 1,524.12 crores, and comprising 17.7% of the quantity and 18.2% of the value of marine product exports from India (KSIDC, 2013). Its seafood industry has continued to grow, being dominated by the export of shrimp, cuttlefish, squid and finfish varieties. In addition to being a source of cheap, nutritious food, the fisheries sector has been recognized as a powerful income and employment generator, and also stimulates the growth of a number of subsidiary industries, representing a major livelihood for a large portion of the low-income population in the country (GOI, 2001). Although fishing is practised along the entire coastal line, intensive fishing activity is centred in Kochi, mainly because of its port facilities for processed fish (Iyer et al., 1994), which contains a vast majority of the export processing plants within the state, with 173 out of the 206 registered exporters in Kerala being situated within the Kochi and Cherthala taluks (MPEDA, 2015).

Water quality can be assessed with regard to physicochemical (Hamid, Masoud, Masoud, & Fardin, 2010) and/or biological analyses (Hillebrand & Sommer, 2000; Sharma & Bhardwaj, 2011). The water transparency in the Carlton trophic state index is a simple parameter for determining aquatic ecosystem health (Rahul, Arvind, & Upadhyay, 2013; Sharma, Kumar, & Rajvanshi, 2010), requiring a minimum of data and generally being easy to understand (Carlson, 1977).

Biomonitoring is a standard method used in conservation studies to assess ecosystem biological integrity (Pai, 2002; Patrick, 1959). The most commonly used method for biological evaluation of the pollution status of aquatic ecosystems focuses on the types, number and abundance of the inhabiting species (Palmer, 1969). The effects of pollutants on the biological community can be considered an early warning sign for such degradation (Walsh, Bahner, & Horning, 1980) with many algae being considered indicators of water pollution (Gunak, 1991; Palmer, 1957; Shotriya & Dubey, 1987). Algal growth depends on sunlight and nutrient concentrations, meaning an excessive level of algae is indicative of nutrient pollution (De Lange, 1994). Algae also are sensitive to some pollutants at levels that might not be evident for other organisms in the short term, or which might affect other communities only at higher concentrations (Mitchell & Stapp, 1992; Relyea, 2005). Initial changes in aquatic communities attributable to increased eutrophication typically begin with successions in the species composition and phytoplankton abundance (Danilov & Ekelund, 1999; Ignatiades, Vassiliou, & Karydis, 1985; Smith, Tilman, & Nekola, 1999), with the dynamic structure of plankton communities dependent on the local environmental conditions (Fernández et al., 1993; Kokuirkina & Mikaelyan, 1994; Linden, Linden, Ganning, & |& Lindestrom, L., 1992; Shushkina & Vinogradov, 1992).

The coastal belt of Vembanad Lake is bordered by a number of seafood processing units with their effluents discharged into the lake system. Apart from the studies on the physicochemical and biological characteristics of the Cochin Estuary, a detailed investigation on this perspective of pollution and its impacts on Vembanad Lake has not yet been conducted, with an integrated approach relating biological and physicochemical aspects with regard to seasons currently not available for this unique backwater ecosystem (Radhika, 2013). Accordingly, the present study has the following objectives:

1. To classify the stretch in the Ramsar site (Vembanad Lake, Kerala) adjacent to the seafood processing units, using Carlson's trophic state index; and
2. To better understand the effects of seafood processing effluents on the physicochemical parameters of the lake and its effects on the phytoplankton population.

## 2 | METHODS

### 2.1 | Study site

The Vembanad Wetland, extending between 09°00′–10°40′N latitude and 76°00′–7°30′ E longitude, is the largest, most important tropical wetland in Kerala, supporting several livelihood activities. This Ramsar site covers an area of 1,513 km$^2$, stretching from Alappuzha to Kochi. Vembanad Lake borders Cherthala, Ambalapuzha and Kuttanad taluks of the Alappuzha District. The present study was conducted on the part of the wetland located in the Cherthala–Aroor–Edakochi coastal belt, the site of a large number of seafood processing plants. It was conducted for 2 years (October 2010–September 2012), comprising ten pre-selected sampling sites ($S_1$–$S_{10}$), including Pattanakkad ($S_1$), Parayakad ($S_2$), Shankaranthodu ($S_3$), Konkeri Bridge ($S_4$), Chandiroor ($S_5$), Edakochi I ($S_6$), Edakochi II ($S_7$), Aroor Mukkam ($S_8$), Arookutty Bridge ($S_9$) and Panavally ($S_{10}$) reference site. Nine of the selected sites are located near seafood processing plant discharge points, while one served as the reference site ($S_{10}$), which was free from seafood plant discharges (Table 1 and Figure 1). The first five sampling sites are situated in interconnected channels, while the remaining sites are in the main waterbody. Sites $S_1$, $S_2$ and $S_3$ constituted the southern part, sites $S_4$ and $S_5$ constituted the central part, and sites $S_6$, $S_7$, $S_8$ and $S_9$ constituted the northern part of the lake. The reference sampling site, located in the main waterbody, was situated in the eastern part of the lake.

### 2.2 | Water quality analyses

Surface water samples from the selected sampling sites were collected during the pre-monsoon (PRM), monsoon (MON) and post-monsoon (POM) seasons. They were collected in plastic bottles, taken to the laboratory and refrigerated at 4°C. The physicochemical parameters, including water and atmospheric temperature and pH, were measured with microprocessor-based portable water quality testing meters. The water salinity was measured with a hand-held refractometer. Alkalinity, free $CO_2$ and BOD were measured and analysed by titration, as recommended by Adoni (1985) and APHA (2005). Nitrate, phosphate, silicate and ammonia–nitrogen concentrations were analysed with an HI 83203 multi-parameter bench photometer for aquaculture (Hanna Instruments Inc., Rhode Island). A 20 m diameter Secchi disc was used to measure water transparency. As there was no significant variation between years, the data of the two-year study (from October 2010 to September 2012) were pooled for three seasons and analysed for seasonal variations with regard to the PRM (February, March, April and May), MON (June, July, August and September) and POM (October, November, December and January) seasons.

The Carlton trophic state index value was calculated with the following formula:

$$\text{Carlton trophic state index (CTSI)} = 10 \times \left(6 - \ln \text{SDD}\right) / \ln 2 \qquad (1)$$

where SDD = Secchi disc reading (cm).

### 2.3 | Phytoplankton analyses

Phytoplankton samples were collected monthly from the surface waters with a conical net of 50 µm mesh size for a period of 2 years. The filtered water was concentrated to 100 ml and preserved in 4% formalin. Taxonomic identification of plankton up to the genus level was done using standard keys (Adoni, 1985; Newell & Newell, 1986; Palmer, 1980; Santhanam, Ramanathan, Venkataramanujam, & Jegatheesan, 1987). Separation and counting of plankton were performed, using 1 ml of subsample in a Sedgwick–Rafter (S-R) plankton counting chamber (1 ml capacity) under a compound microscope (Olympus, Japan).

## 3 | RESULTS AND DISCUSSION

### 3.1 | Variation of trophic state index for different sampling sites

The Carlton trophic state index classifies a lake as oligotrophic (<40: low productivity), mesotrophic (40–50: moderate productivity), eutrophic (50–70: highly productive) or hypereutrophic (>70). The index values for different sampling sites exhibited seasonal fluctuations in the present study.

The season-wise average of the Carlton trophic state index (CTSI) for the ten sites varied from 64.5 to 81.1 during PRM, 62.8

**TABLE 1** Characteristics of study sites

| Site number | Name | Geographical position | Source of pollution | Land utilization | Colour | Remarks |
|---|---|---|---|---|---|---|
| 1 | Pattanakkad | 9°44′22N, 76°19′07E | Discharge from peeling shed | Panchayat area Residential | Turbid water | Interconnection canal |
| 2 | Parayakad | 9°47′13N, 76°18′20E | Discharge from seafood processing industry | Aquaculture farm | Turbid water | Interconnection canal |
| 3 | Shankaranthodu | 9°47′22N, 76°18′12E | Discharge from peeling shed | Panchayat area Residential | Highly turbid water | Interconnection canal |
| 4 | Konkeri Bridge | 9°49′22N, 76°18′22E | Discharge from seafood processing industry | Panchayat area Residential | Turbid water | Interconnection canal |
| 5 | Chandiroor | 9°50′28N, 76°18′29E | Discharge from seafood processing industry | Panchayat area Residential | Highly turbid water | Interconnection canal |
| 6 | Edakochi I | 9°54′47N, 76°17′06E | Discharge from seafood processing industry | Panchayat area Residential | Highly turbid water | Main waterbody |
| 7 | Edakochi II | 9°54′07N, 76°17′42E | Discharge from seafood processing industry | Boat building yard | Highly turbid water | Main waterbody |
| 8 | Aroor Mukkam | 9°53′23N, 76°17′49E | Discharge from seafood processing industry | Fallow land | Turbid water | Main waterbody |
| 9 | Arookutty | 9°52′12N, 76°19′01E | Discharge from seafood processing industry | Panchayat area Residential | Highly turbid water | Main waterbody |
| 10 | Panavally/(Reference site) | 9°49′13N, 76°21′33E | Free from seafood processing waste discharges | Agriculture and Aquaculture | Clear water | Main waterbody |

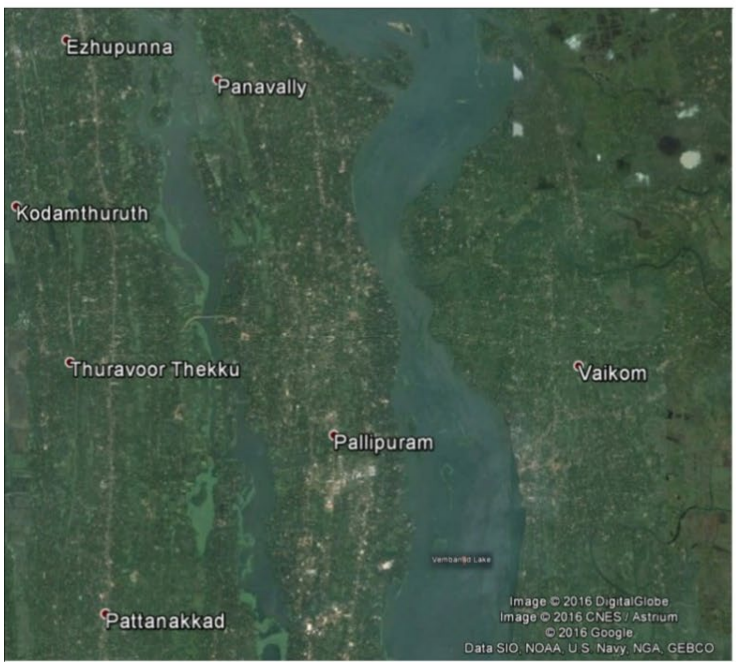




**FIGURE 1** Study site

to 78.4 during the MON, and 62.6 to 78.9 during the POM seasons, indicating the CTSI was maximum during the PRM season and minimum during the MON season (Figure 2). The reference site ($S_{10}$) exhibited the lowest CTSI value during all seasons.

A hypereutrophic state was observed for the sites in the interconnected channels of $S_3$ and $S_5$, and in the main waterbody ($S_9$), which were located close to the seafood processing units/peeling shed discharge points, confirming these effluents were mainly responsible for the degraded water quality. Hypereutrophication was of the greatest magnitudes in the interconnected channel ($S_5$) during PRM and MON seasons (near the seafood processing unit) and at site $S_3$ near the peeling shed during the POM season, indicating the interconnected channels are mostly affected by the effluent because of little dilution possibilities, noting heavily turbid water exists in these areas.

The variation of CTSI from 2010 to 2011 is shown in Figure 3. Eutrophic conditions exist in the lake in all sites, except for $S_2$, $S_5$ and $S_9$ during the MON season. During other seasons, hypereutrophic conditions characterize all the sampling sites, except for the reference site ($S_{10}$), indicating heavy rainfall during the MON season had resulted in some improvement in the water quality. The sites most affected are in the interconnected channels ($S_1$, $S_3$, and $S_5$) and in the main waterbody ($S_9$).

The variation of CTSI from 2011 to 2012 is shown in Figure 4. During the PRM and MON seasons, hypereutrophic conditions exist at all the sites, except for the reference site ($S_{10}$). This indicates more severe water quality deterioration occurred in 2011–2012 than in 2010–2011. Hypereutrophic conditions also exist at all the sites, except for $S_4$, $S_6$, $S_8$ and $S_{10}$ during the POM season. The sites most affected are in the interconnected channels, namely the central part ($S_5$) and the northern part of the main waterbody ($S_7$ and $S_9$). Thus, the decreased rainfall affected the water quality of the main waterbody more than that of the interconnected channels.

## 3.2 | Seasonal water quality variations

### 3.2.1 | Temperature

Temperature plays a major role in the physicochemical and biological behaviour of water ecosystems (Dwivedi & Pandey, 2002; Welch, 1952). The atmospheric temperature exhibited significant variation among sampling sites and seasons, whereas the water temperature exhibited significant variations between seasons. The difference in the water temperature might be attributable to the sampling time and seasonal influence, as suggested by Desai (1995) and Jayaraman, Devi, and Nayar (2003).

The average atmospheric temperature was 30.7, 33.8 and 31.3°C during the PRM, MON and POM seasons, respectively, from 2010 to 2011. The lower value was observed in the southern portion of the waterbody (i.e. interconnected channel $S_1$) during the PRM (28.8°C), MON (32.3°C) and POM (29°C) seasons. The high temperature was observed for the reference site in the main waterbody ($S_{10}$) during the PRM (31.8°C) and in the northern part of the main waterbody ($S_7$) during the MON (34.8°C) and POM (33°C) seasons. The increased atmospheric temperature enhanced the evaporation rate, resulting in reduced water depth.

The average water temperature was 28.8, 31.5 and 30.5°C during the PRM, MON and POM seasons, respectively, from 2011 to 2012. The lower temperature was observed in the southern portion (i.e. interconnected channel) during the PRM and POM seasons, while the higher temperature was observed in the northern portion of the lake (i.e. in the main waterbody). The water temperature was high in the main waterbody and low in the interconnected channels.

The water temperature and atmospheric temperature did not exhibit much variation in the present study, with the recorded values being in a narrow range. Studies conducted in the retting zones by Remani (1979) in Cochin backwaters, Aziz and Nair (1986) in the Edava-Nadayara Estuary and in the Kadinamkulam-Anchuthengu estuarine system (Nandan, 1991; Nandan & Azis, 1994) demonstrated higher water temperatures in the retting zones than in the non-retting zones. In contrast, the water temperature did not exhibit such differences between the polluted sampling sites and the reference site in the present study.

### 3.2.2 | pH

The pH of water can provide an indication of the intensity of water pollution (Verma & Shukla, 1970). The average pH value was 7.20, 7.60 and 7.85 during the PRM, MON and POM season, respectively, in the present study.

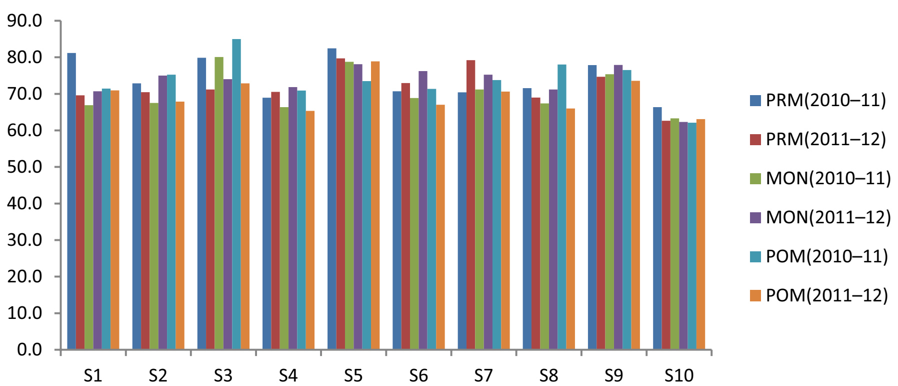



**FIGURE 2** Variation of Carlson trophic state index from 2010 to 2011 and from 2011 to 2012

**FIGURE 3** Variation of Carlson trophic state index from 2010 to 2011

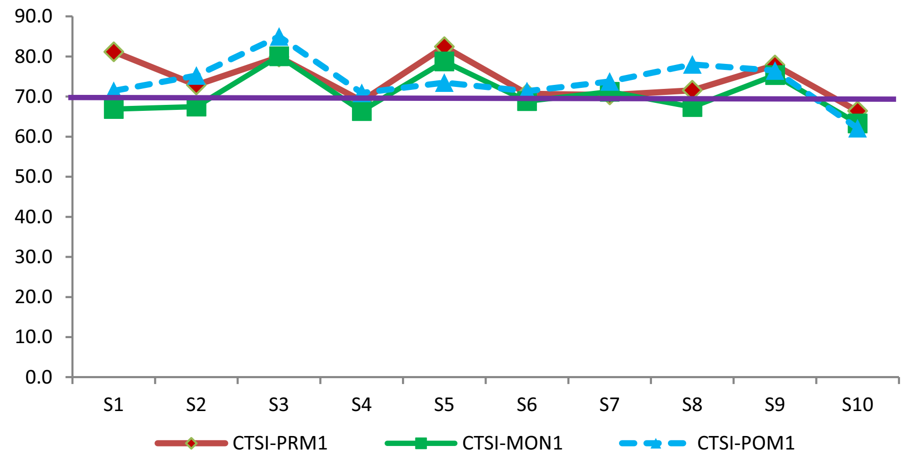
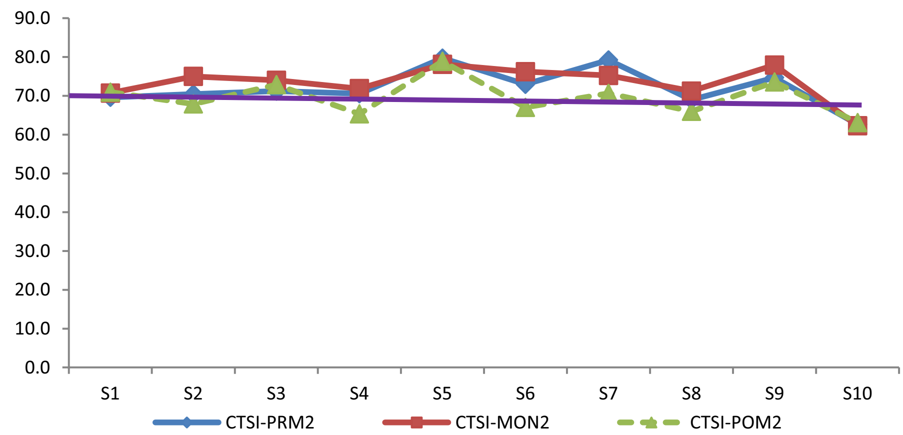



**FIGURE 4** Variation of Carlson trophic state index from 2011 to 2012




The lowest pH was observed at sampling site $S_5$ in the PRM (6.69) and MON (7.21) seasons, and at site $S_8$ (7.32). The lower pH might be attributable to decomposition of the organic matter entering the waterbody. The highest pH was observed in the interconnected channel ($S_1$) during the PRM (7.91) and POM seasons, and at site $S_2$ (8.08) during the MON season. The pH during the present study indicated an alkaline condition, exhibiting significant variations between sampling sites and seasonally. The pH in the interconnected channels ($S_1$–$S_2$) was high, compared to the main waterbody and reference site ($S_{10}$). The low pH at the northern sites ($S_6$–$S_9$) in the main waterbody might be attributable to the waves and its connection to the sea, which diluted the waste upon its discharge. In examining the physicochemical characteristics of seafood effluents from Aroor Grama Panchayath, Thomas, Nair, and Singh (2015) reported the pH varied from 6.8 to 7.5. Effluents from fish processing establishments are seldom acidic, being close to a pH value of 7.0 or alkaline (Alex, 2005; Chowdhury, Viraraghavan, & Srinivasan, 2010; Sankpal & Naikwade, 2012; Thomas et al., 2015; Vaghela, Krishnakumar, & Dar, 2015), thought to be attributable to decomposition of proteinaceous matter in the effluent (Alex, 2005). If the current situation of waste disposal into the lake continues, the water quality will shift to an alkaline range over time, affecting the aquatic life, noting either highly acidic or alkaline water can kill marine life (Lokhande, Singare, & Pimple, 2011).

### 3.2.3 | Salinity

Salinity also has a major role in controlling various physical, chemical, and biological processes in aquatic environments. Salinity can indicate seawater intrusion or pollution attributable to sewage and/or industrial wastes. The average salinity value was 9.0, 4.3 and 9.2 ppt during the PRM, MON and POM season, respectively, with the lower value observed during the MON season (4.3 ppt), whereas the maximum value was observed in the POM season(9.2 ppt). The salinity was low at the reference site in the eastern part of the wetland ($S_{10}$) during the PRM (4.88 ppt) and MON (1.38 ppt) seasons, and in the southernmost part of the interconnected channel ($S_1$) during the POM season. The lower salinity occurred at these sites because of their location away from the estuary mouth. The high salinity level was observed in the northern part ($S_8$) during the PRM (12.38 ppt), MON (8.38 ppt) and POM (11.2 ppt) seasons. The nearness of these sites to the sea might be the reason for the high salinity values. The average salinity value was 7.5 ppt.

### 3.2.4 | Free carbon dioxide ($CO_2$)

The main source of $CO_2$ is from the atmosphere, from respiration of animals and plants, and from bacterial decomposition of organic matter. The average free $CO_2$ value was 45.82, 76.04 and 53.50 mg/L during the PRM, MON and POM seasons, respectively. The lower free $CO_2$ value was observed during the PRM season (45.82 mg/L) and the higher value during the MON season (76.04 mg/L). In comparing the sampling sites, the free $CO_2$ was low at the eastern reference sampling site ($S_{10}$) during all seasons, reflecting a smaller load of organic matter in the water, as reported by Pathani and Upadhyay (2006). The alkaline nature of water is mainly responsible for the generation of free $CO_2$. High values were obtained for the processing effluent-affected central part of the lake ($S_5$) during the PRM (130.88 mg/L) and MON seasons (163.00 mg/L), and at site $S_9$ during the POM season (73.13 mg/L). The present study revealed that the interconnected channels are more affected than the main waterbody. The average free $CO_2$ value observed during the study period was 58.45 mg/L, with the fish processing effluent resulting in the increased free $CO_2$ value in this part of the wetland.

### 3.2.5 | Alkalinity

Alkalinity indicates the presence of the salts of weak acids such as carbonates, bicarbonates, phosphates and silicates together with free hydroxyl ions in solution. The input of wastes to waterbodies can also increase alkalinity levels (Mulani, Mule, & Patil, 2009). The average alkalinity was 111.55, 98.01 and 80.64 mg/L during the PRM, MON and PRM seasons, respectively. The lower value was reported during the POM season (80.64 mg/L) and the highest value during the PRM season (111.55 mg/L). The alkalinity was low at the eastern reference sampling site ($S_{10}$) during all seasons, indicating a smaller pollution load, while it was high in the central part of the waterbody ($S_5$) during the PRM (234.38 mg/L) and MON seasons (197.63 mg/L), and in the northern part ($S_9$) during the POM season (130.75 mg/L). Alkalinity values exceeding 100 mg/L are classified as eutrophic, while those less than 50 mg/L are classified as oligotrophic (Anon, 2001). Thus, the central part of the wetland ($S_3$, $S_5$ and $S_9$) can be classified as eutrophic, while the reference site ($S_{10}$) can be considered oligotrophic. The average alkalinity value during the study period was 96.73 mg/L. The increased alkalinity is attributable to the discharge of untreated processing effluents into the wetland. The alkalinity exceeded the permissible limit of 200 mg/L, as described by BIS, in the central portion of the waterbody ($S_5$).

### 3.2.6 | BOD

The biochemical oxygen demand (BOD) is the quantity of oxygen required by microorganisms to decompose biologically degradable organic matter in water under aerobic conditions. The average BOD value during the study period was 28.01, 30.81 and 29.59 mg/L during the PRM, MON and POM seasons, respectively (Figure A1). The lower value was observed during the PRM (28.01 mg/L) and the higher value during the MON season (30.81 mg/L). The BOD was low at the east reference sampling site ($S_{10}$) during all seasons, indicating less organic pollution at this site. The highest BOD value was observed in the central portion during the PRM (42.70 mg/L), MON (S2:45.61 mg/L) and POM seasons (S5:38.66 mg/L). The average BOD value was 29.47 mg/L. Based on primary water quality criteria for bathing water, as prescribed by the Central Pollution Control Board, the BOD should not exceed 3 mg/L. The average BOD level, however, was more than nine times higher than the standard value, clearly indicating a high BOD level in this part of the wetland.

### 3.2.7 | Phosphate

Phosphorous is often regarded as the most important algal-limiting nutrient in freshwater systems, with the phosphorus concentration greatly influencing the abundance of plankton. The average value of phosphate was 2.15, 2.97 and 2.28 mg/L during the PRM, MON and POM seasons, respectively. The lower value of phosphate was observed in the PRM season (2.15 mg/L), while the higher value was observed in the MON season (2.97 mg/L). The lower phosphate value was observed at the eastern reference site during all seasons, with the higher values occurring in the northern portion ($S_9$) during the PRM (4.04 mg/L), in the central portion ($S_5$) during the MON (10.51 mg/L), and in the southern part ($S_3$) during the POM season (2.84 mg/L). A high phosphate concentration was observed in the interconnected channels, confirming the polluted nature being attributable to untreated waste discharges. The higher level in the MON season might be attributable to surface run-off carrying untreated effluents. Hutchinson (1957), Welch (1952) and Ruttner (1953) reported very little phosphate in undisturbed natural waters, in accordance with the investigations of Venkateswarlu (1969a, 1969b) and Amarendra Kumar (2010). The average phosphate value was 2.47 mg/L.

### 3.2.8 | Nitrate

Nitrate has an important role in balancing various biological processes occurring in the aquatic ecosystem. The average nitrate concentrations were 11.03, 9.19 and 8.52 mg/L during the PRM, MON and POM seasons, respectively (Figure A2). The lower nitrate values were noted during the POM (8.52 mg/L) and higher in the PRM season (11.03 mg/L). The nitrate concentration was low at the reference site and high in the northern part of the lake ($S_9$) during all seasons. It was high in the main lake waterbody, indicating the addition of processing effluents into the wetland system, with nitrate indicating the stabilization of organic waste. Furthermore, to ensure the quality and food safety of the finished products, the industry uses a variety of chemicals (e.g. food additives, cleaners, disinfectants) which can also become an important organic pollution source, since these additives might contain nutrients, such as phosphorus (P) and nitrogen (N), which eventually end up in the effluents, thereby contributing to the pollution and eutrophication of coastal waters (Lalonde, 2009; Morry et al., 2003; Tchoukanova et al., 2003). Inorganic nutrients, such as water-soluble nitrogen and phosphorus, can also cause excessive algal growths (Andrew & Jakson, 1996; Miller, 1995; Waite, 1984). The average nitrate concentration during the study period was 9.58 mg/L.

### 3.2.9 | Ammonia–nitrogen

The content of ammonia and ammonium ions in water serves as an indicator of water pollution. The seasonal and site-wise variations of this parameter are illustrated in Figure A3. The average ammonia–nitrogen concentration was 2.82, 5.36 and 4.59 mg/L during the PRM, MON and POM seasons, respectively. The lower value was noted during the PRM (2.82 mg/L) and higher value in the MON season (5.36 mg/L). The ammonia–nitrogen concentration was low at the reference site, with a higher value at the central sampling site ($S_5$) during the entire study period. The average ammonia–nitrogen concentration was 4.26 mg/L. The high content of ammonia–nitrogen in seafood processing effluents is the main source for its increased level in the wetland system. The decay process of fish and shellfish also can release significant quantities of ammonia and nitrate (Leffler, 1997).

### 3.2.10 | Silica

The average value of silica was 2.77, 3.88 and 1.93 mg/L during the PRM, MON and POM seasons, respectively. The lower silica concentration was observed during the POM (1.94 mg/L) and the high in the MON season (3.88 mg/L). According to Livingstone (1963), the world-wide mean concentration of silicates is 13.0 mg/L. The average concentration in the present study did not exceed 4.60 mg/L, possibly attributable to its utilization by diatoms. The lower value was observed at the reference site during all seasons, and the higher value was observed at the northern site ($S_9$) during the PRM season (4.18 mg/L), while the high values were observed in the MON and POM seasons at the central site ($S_5$). The higher silica concentration is an indication of waste from industrial effluents, as well as contributions from the death of diatoms. The average silica concentration was 2.86 mg/L.

## 3.3 | Phytoplankton

A total of 181 genera numbering 38,472 phytoplankton individuals belonging to 11 classes were observed from all the sampling sites during the present study (Table A1). A total of 21,812 individuals represented class Bacillariophyceae, while class Chlorophyceae and Cyanophyceae were represented by 7,111 and 6,118 individuals, respectively. The class Bacillariophyceae was the most dominant class, while class Chlorophyceae was the dominant genera. The next dominant class was Dinophyceae (2,445 individuals) followed by Euglenophyceae (511 individuals) and Eustigmatophyceae (181 individuals). The class Rhodophyceae was represented by 92 individuals, whereas the class Haptophyceae contained 79 individuals. The class Chrysophyceae (71 individuals) was followed by Cryptophyceae (30 individuals) and Dictyochophyceae (22 individuals). The most dominant classes are discussed in the following section.

### 3.3.1 | Bacillariophyceae

The qualitative and quantitative dominance of diatoms in an aquatic ecosystem is a major indicator of water quality and environmental conditions since they are adapted to a wide range of physicochemical parameters (Ajuonu et al., 2011). The average number of Bacillariophyceae observed was 79, 83 and 111 during the PRM, MONand POMseasons, respectively. The occurrence of Bacillariophyceae was comparatively lower in the PRM and higher in the POM season. The lowest number was observed at the northern polluted site ($S_9$) in the PRM season (54) and in the less-polluted reference site ($S_{10}$) during the MON (42) and POMseasons (44). The highest number was reported in the southern sites (114 at $S_1$ (PRM); and 148 and 155 at $S_3$ (MON and POM)). Not all the members contributed to the dominance of different classes of plankton. At the eutrophic sampling sites, a few indicator species comprised a large portion of the different plankton classes. Nandan and Aher (2005) have reported that diatoms (*Nitzschia*, *Navicula*, etc.) are found in organically polluted water. In this part of the lake, organic pollution indicators (*Nitzschia* sp.; *Navicula* sp.) are abundant. The high number of Bacillariophyceae in all seasons indirectly indicates surplus nutrients in the waterbody. The present study supports the findings of Huang et al. (2004), who suggested these eurythermal phytoplankton species grow quickly under eutrophic conditions, with Bacillariophyceae occupying the first position, followed by Chlorophyceae. Diatoms are the best pollution indicators, having one of the shortest generation times of all biological indicators, reproducing and responding rapidly to environmental changes, thereby being early warning indicators of both pollution and habitat restoration needs (Stoermer & Smol, 1999).

### 3.3.2 | Dinophyceae

Dinophyceae is the dominant component of photosynthetic organisms. The average number of Dinophyceae observed was 8, 8 and 13 during PRM, MON and POM seasons, respectively. The lower number was observed during the PRM and MON seasons (8) and higher in the POM season (13). The lowest number was present in the less-polluted reference site during all seasons. Dinoflagellates usually prefer oligotrophic waters, being absent in eutrophic waters because of competition with diatoms (Cushing, 1989; Menzel, Hulbert, & Ryther, 1963). There was no Dinophyceae representative found in the central part of the lake ($S_4$) during the MON season. The highest number was observed at the southern site ($S_2$) during the PRM (12), at $S_3$ during the MON (26) and at the northern sampling site ($S_9$) during the POM season (24). Even though their number was high, they cannot outnumber the diatoms under eutrophic conditions. The dominant member in this class was *Ceratium* sp., which is known to proliferate in nutrient-rich waters (Cander-Lund & Lund, 1995). In estuarine conditions, dinoflagellates represent an important plankton component in polluted water, whereas they are practically absent in polluted freshwater areas (Palmer, 1980). The average number observed during the present study was 10.

#### 3.3.3 | Chlorophyceae

They are an abundant phytoplankton group because of their tolerance to a wide range of physicochemical parameters. The average value of Chlorophyceae was 27, 24 and 39 during the PRM, MON and POM seasons, respectively. The lower number was observed in the MON (23), while higher number was observed in the POM season (39). The lower number was observed at the less-polluted reference site (S10) during all seasons. The highest number was observed at the southern site ($S_2$) during the PRM (47) and POM (94) seasons and at site $S_3$ during the MON season (40). The dominant members of this class were *Ankistrodesmus* sp and *Scenedesmus* sp. The presence of these genera indicated a eutrophic environment (Tripathi, Pandey, & Tiwari, 1987; Zargar & Gosh, 2006). Like Bacillariophyceae, their abundance is an indicative of large nutrient loads, regardless of the season. The average number of Chlorophyceae observed during the study period was 30.

#### 3.3.4 | Cyanophyceae

The presence of blue-green algae, as the most abundant genera, indicates the water is not potable unless properly treated prior to use. Their dark blue-green algal blooms are indicative of eutrophication. The average number of Cyanophyceae was 30, 25 and 21 during the PRM, MON and POM seasons, respectively. The lower number was observed in the POM (21), and the higher in the PRM season (30). Epilithic algae such as *Oscillatoria* sp and *Phormidium* sp were higher in number, indicating the presence of pollutants of biological origin, and being excellent indicators of water pollution (Round, 1965). The blue-green algal population was high in the PRM season, possible attributable to elevated water temperature and the presence of high organic matter content. The population was low at the less-polluted reference site (S10) during all seasons. The high number was observed in the southern part of the lake ($S_2$) during the PRM season (83), at $S_3$ during the MON season (45) and in the central part ($S_5$) during the POM season (49). The high abundance of Cyanophyceae, irrespective of season, is an indication of eutrophication. The average number observed during the present study was 25. Brinley (1942), and Venkateswarlu (1969b) and Sampath Kumar (1977) reported that blue-green algae are often profuse in highly polluted habitats, with Franklin (1972) suggesting blue-green algae are general indicators of eutrophication, with their growth increasing with increasing organic matter.

Pollution-denoting parameters (e.g. BOD, alkalinity, ammonia–nitrogen, phosphate, silica) were observed less often in the eastern part ($S_{10}$), which does not receive seafood wastes. In contrast, their concentrations are high in the southern, central and northern portion of the lake. Phytoplankton growth and development are a function of such physicochemical parameters, with the basic process of phytoplankton production also dependent on temperature, turbidity and nutrients (Sreenivasan, Sampathy, & Paramasivam, 1979; Sukumaran & Das, 2002). One of the major selective environmental factors restricting species numbers is high water salinity (Qasim, Wellershaus, Battathiri, & Abidi, 1969; Soyalu & Gonulol, 2010), with higher alkalinity, nitrate, ammonia–nitrogen and phosphate favouring enrichment of Cyanophyceae (Jarousha, 2002; Padmavathi & Veeraiah, 2009).

### 3.4 | Relationships between water quality and dominant phytoplankton classes

Physicochemical parameters influence the distribution of certain species of plankton and vice versa (Sharma & Singh, 2013). Accordingly, correlations between biotic and abiotic factors are useful in assessing the trophic status of a waterbody (Sharma, Parashar, Bagre, & Qayoom, 2015). To this end, correlation tendencies of phytoplankton density with different physicochemical water quality parameters were not identical at all sampling sites in the present study (Table A2). Gaur and Kumar (1981) reported that growth regulators in the effluents in specific concentrations can enhance phytoplankton growth rates. Correlation analysis of the density of different algal classes and temperature indicated differences in the interconnected water channels and in the main waterbody. For site 5, Cyanophyceae exhibited a significant negative correlation with water temperature. Palmer (1980) reported the optimum temperature for diatoms as 18–30°C, for green algae 30–35°C, and for blue-green algae 35–40°C. The temperature ranged from 28 to 32°C in the present study, not being favourable for Cyanophyceae. Bacillariophyceae exhibited a significant negative correlation with pH, ammonia–nitrogen and silica. Diatoms require silica in soluble forms for wall silicification, leaving less silica present in the water (Reynolds, 1984). As silicate is the main nutrient in diatom metabolism, its abundance enhances the multiplication of diatoms, noting that natural waters exhibit a decline in dissolved silica concentration during times of maximum diatom growth (Boney, 1989). Many investigators stressed the importance of silicate in the periodicity of Bacillariophyceae (Eggs & Aksnes, 1992; Krishnamurthy & Bharti, 1996; Millman & Boyle, 1975). The pH affects the carbonate–bicarbonate buffering system, noting that carbon available for diatom growth is in the form of $CO_2$ or $HCO_3^-$ at low pH values, whereas it is in the form of bicarbonate and carbonate at high pH values (Penny, 1993). The diatoms might use the hydrogen ions for growth, resulting in their negative correlation observed in the present study. The low pH was reported at the $S_5$ site, favouring the growth of *Coscinodiscus* sp., *Fragillaria* sp., *Navicula* sp. and *Nitzchia* sp. Maheshwari (2011) reported *Coscinodiscus oculus* and *Fragillaria* *intermedia* were adapted to grow at lower pH values. Yoshiyama and Sharp (2006) suggested ammonia–nitrogen can exert a strong negative influence on phytoplankton production above a relatively low concentration. As nitrate is an ammonia source in water, nitrate might be used by plankton for growth before its oxidation to ammonia (Rajasegar, 2003). The reported higher concentration could be partially attributed to the death and subsequent decomposition of phytoplankton (Segar & Hariharan, 1989), being a possible reason for their negative relationship. Chlorophyceae exhibited a significant positive correlation with salinity, nitrate and phosphate, and a significant negative correlation with pH and free $CO_2$. The phytoplankton exhibited a direct positive relationship with phosphate, indicating higher phosphate concentration enhanced their growth (Krishnan et al., 1999). The dissolved solids mainly comprise carbonates, bicarbonates, chloride, sulphate, calcium, magnesium, phosphate, nitrate, sodium, potassium and iron (Trivedy and Goel, 1986). An increase in the nitrate concentration promotes phytoplankton growth (Nandan & Patel, 1992), which might be the reason for its positive relationship with Chlorophyceae. The high organic matter in the seafood effluent discharges might be the reason for the higher total dissolved solids at the polluted sampling sites. Cyanophyceae exhibited a significant negative correlation with alkalinity and water temperature. Furthermore, higher hardness levels in the interconnected channels, irrespective of season, indicate eutrophication, favouring the growth of pollution-tolerant Cyanophyceae. In contrast, the pollution-sensitive Cyanophyceae decreased with increasing alkalinity. The death and decay of plankton might release nutrients, increasing the alkalinity, noting similar observations were reported for a pond ecosystem (Duttagupta, Gupta, & Gupta, 2004).

The pH exhibited a significant positive correlation with Cyanophyceae at polluted sampling sites in the main waterbody, whereas it exhibited a significant negative correlation with Bacillariophyceae (Table A3). This is in contrast to the results of Bhatt, Lacoul, Lekhal, and Jha (1999), who reported a negative correlation coefficient with Cyanophyceae and Chlorophyceae, and a positive one with Bacillariophyceae. According to Palmer (1980), most algae grow best in water at or near a neutral pH level, although Cyanophyceae grow best at a high pH. A high alkaline pH was observed in the interconnected channels, especially at sites $S_1$ and $S_2$, which were also the sites with the highest number of individuals of Cyanophyceae (715 and 977). The high BOD in the interconnected channels at site $S_7$ favoured the growth of Dinophyceae. A high BOD concentration can result from the decomposition of the organic matter, serving as the food for increased algal populations (Jamil, 2001). The negative interaction between BOD and Cyanophyceae at site $S_9$ might be attributable to unidentified interactions of different factors operating in that specific site. Brinley (1942), and Venkateswarlu (1969a, 1969b) reported blue-green algae occur in large numbers in polluted habitats. Oviatt, Lane, French, and Donaghay (1989) and Wladyslawa, Agnieszka, and Michał (2007) related the abundance of phytoplankton, particularly blue-green algae, to increased nutrient loads. In the present study, however, Chlorophyceae exhibited a significant positive correlation with nitrate at site $S_{10}$. Harish (2002) concluded that phosphates, nitrates and nitrites control Chlorophyceae growth, with increased concentrations of nitrates promoting phytoplankton growth (Nandan & Patel, 1992).

#### 3.4.1 | Relationships between characteristics of selected raw effluents and dominant phytoplankton and zooplankton classes in the interconnected channels

A correlation analysis was done to better understand the influence of raw effluents on the dominant phytoplankton classes in the interconnected channels and in the main waterbody (Table A3). The effluent characteristics were obtained from earlier research in the Alappuzha District (Sheeba, 2008). In the interconnected channels, the pH negatively influenced the Bacillariophyceae, Dinophyceae, Chlorophyceae, Cyanophyceae, Cladocera and Copepoda populations, whereas the raw effluent positively influenced the Copepod plankton population. The BOD, nitrate and ammonia–nitrogen in the raw effluent positively influenced Bacillariophyceae, Dinophyceae, Chlorophyceae, Cyanophyceae, Cladocera and Copepoda, whereas they negatively influenced the Tintinnid population. Bacillariophyceae, Dinophyceae and Chlorophyceae positively favoured each other and Copepoda, Cladocera and Rotifera, except for the tintinnids.

#### 3.4.2 | Assessment of relationships between selected raw effluent characteristics with dominant phytoplankton and zooplankton classes in the main waterbody

In the main waterbody, the pH negatively influenced BOD, nitrate, ammonia–nitrogen, Cyanophyceae and Copepoda, while it positively influenced the Tintinnids and Cladocera. The BOD positively favoured the Cyanophyceae and Copepoda, while it negatively favoured Bacillariophyceae, Dinophyceae, Chlorophyceae, Tintinnids, Cladocera and Rotifera. Nitrate positively affected the phytoplankton and zooplankton classes, while ammonia–nitrogen negatively influenced the Chlorophyceae, Tintinnid and Cladocera. The remaining classes exhibited a positive correlation with ammonia–nitrogen. Bacillariophyceae and Dinophyceae positively favoured each other, and other classes of phytoplankton and zooplankton. Chlorophyceae positively favoured Tintinnids, Cladocera and Rotifera, while Cyanophyceae positively influenced Copepoda.

## 4 | CONCLUSIONS

The present study was undertaken to assess threats to a Ramsar wetland from fish processing waste effluents. The factors responsible for eutrophication were identified for different stretches of the lake system. The study results indicated hypereutrophic conditions existed in that part of the wetland near the effluent discharge points. Analysis of phytoplankton communities supported the interpretation of the results based on the physicochemical analysis of the water and vice versa. Hypereutrophic conditions were observed more in the interconnected channels than in the main waterbody. Free carbon dioxide, BOD phosphate, ammonia–nitrogen and silicate were found to exhibit high levels during the MON season. High concentrations of alkalinity and nitrate, as well as a high Cyanophyceae population, occurred during the PRM season. High salinity and low alkalinity and nitrate conditions, and high populations of Bacillariophyceae, Dinophyceae and Chlorophyceae were observed during the POM season. High levels of free $CO_2$, BOD, phosphate, nitrate and ammonia–nitrogen in the wetland confirm the degradation of the lake water attributable to the fish processing waste effluents.

The nutrients in the interconnected channels, especially ammonia–nitrogen and nitrate, contributed to high CTSI values, whereas organic matter was the predominant contributor to the elevated CTSI in the main waterbody. The high number of Bacillariophyceae during all seasons indirectly indicates surplus nutrients in the waterbody. Similar to Bacillariophyceae, the abundance Chlorophyceae is indicative of a large nutrient load, irrespective of the season. The excess level of organic matter, phosphate and ammonia–nitrogen might have contributed to the increased Cyanophyceae numbers, with anaerobic condition also favouring the occurrence of Cyanophyceae. Bacillariophyceae, Chlorophyceae, Cyanophyceae and Dinophyceae are responsible for the increased trophic state index value for the main waterbody. Unlike the interconnected channels, Cyanophyceae was associated with undecomposed organic matter in the main waterbody.

The occurrence of Bacillariophyceae, Dinophyceae, Chlorophyceae and Cyanophyceae is high where the organic matter (BOD), phosphate, nitrate and ammonia–nitrogen levels are high. A reduced salinity results in reduced Dinophyceae, Chlorophyceae and Cyanophyceae populations, whereas increased nitrate and ammonia–nitrogen levels favour the growth of Bacillariophyceae.

Organic matter (BOD) plays an important role in eutrophication in the central portion of the waterbody, particularly in the interconnected channels, with the stagnant nature of the lake possibly exacerbating the situation. A few indicator species compose a large portion of the different classes of plankton at the sampling sites that exhibit eutrophic conditions, with the dominance of Cyanophyceae over Chlorophyceae, especially in the interconnected channels, is a defining characteristic of polluted water.

The seasonal influence on the correlations between different parameters was consistent throughout the lake. There was a strong positive correlation between BOD and the nutrients (ammonia–nitrogen, phosphate, nitrate, silica) with eutrophic conditions as defined by the Carlson trophic status index (CTSI). High Chlorophyceae numbers observed during the POM season might be attributable to high DO and bicarbonate concentrations observed during these periods, favouring its quick growth. The highest Cyanophyceae count was observed in the MON season, possibly attributable to the low oxygen and high organic matter concentrations. Various phytoplankton classes (Bacillariophyceae, Dinophyceae, Chlorophyceae and Cyanophyceae) contributed to the high CTSI values, irrespective of the seasons.

There was a dominance of Bacillariophyceae, followed by Chlorophyceae, Cyanophyceae and Dinophyceae. Bacillariophyceae, Chlorophyceae and Cyanophyceae were comparatively higher in the interconnected channels, whereas Dinophyceae was high in the main waterbody. There is a shift in dominance from Chlorophyceae to Cyanophyceae in the interconnected channels, especially in the southern and central portion of the lake. The dominance of Cyanophyceae over Chlorophyceae, especially in the interconnected channels, is indicative of polluted water.

Not all plankton species contributed to the dominance of the different plankton classes. The indicator species comprised a large portion of the different plankton classes at the sampling sites exhibiting eutrophic conditions. The main organic pollution indicators (*Nitzschia* sp.; *Navicula* sp.) were found abundant in the class Bacillariophyceae. The dominant member in the class Dinophyceae was *Ceratium* sp, which is known to proliferate in nutrient-rich waters. The dominant members of the class Chlorophyceae were *Ankistrodesmus* sp. and *Scenedesmus* sp. Epilithic algae (e.g. *Oscillatoria*; *Phormidium*) exhibited higher numbers in the class Cyanophyceae, indicating the presence of pollutants of biological origin.

Most of the phytoplankton species observed in the present study are considered to be pollution indicator species. Their increase in water might cause allergic skin diseases, gastrointestinal problems and even death of vertebrates. The waste discharges from the seafood processing industry is a major reason for the increasing organic pollution and eutrophication. A persisting situation of this type will definitely degrade the water quality, with likely negative impacts on aquatic organisms in the lake, including fishes which, ironically, will also adversely affect the seafood industry. The results of the present study suggest it would be undesirable for further expansion of the seafood industry in the Cherthala–Aroor–Edakochi coastal belt of Vembanad Lake.

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## APPENDIX

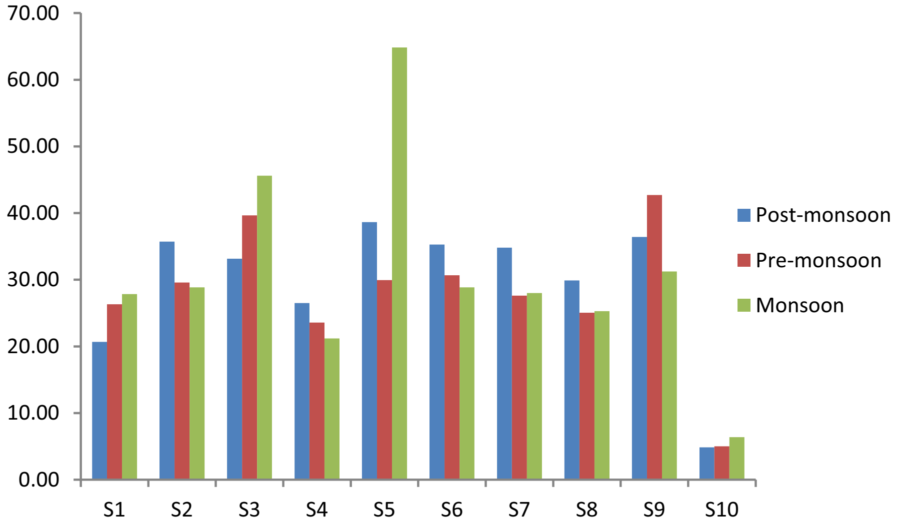


**FIGURE A1** Site-wise and season-wise variation of biochemical oxygen demand (BOD)

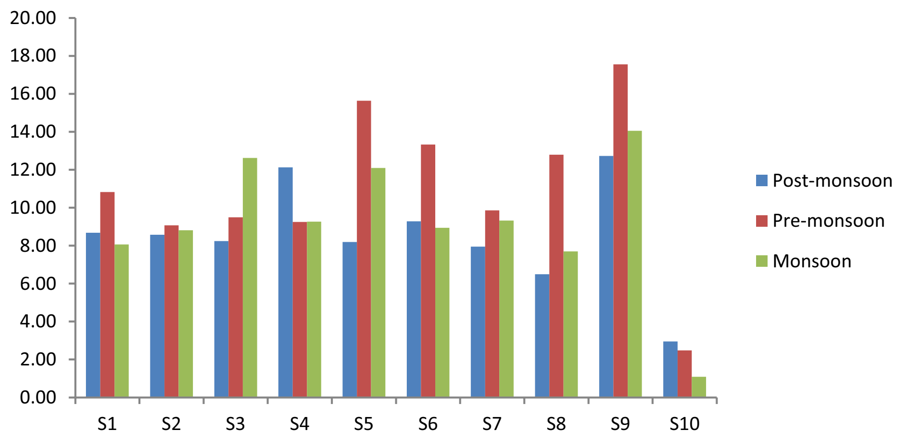


**FIGURE A2** Site-wise and season-wise variation of nitrate

**FIGURE A3** Site-wise and season-wise variation of ammonia–nitrogen

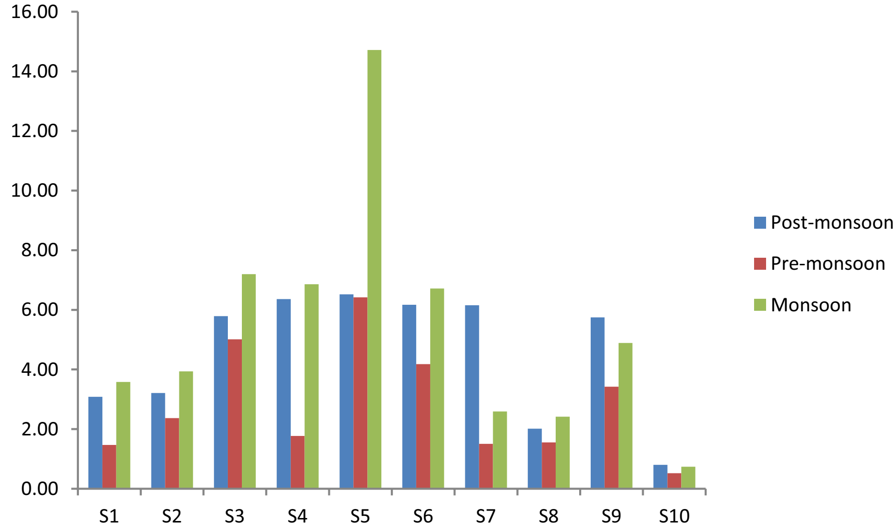


**TABLE A1** Distribution of phytoplankton class for ten sampling sites (#/ml)

| Phytoplankton sp | S1 | S2 | S3 | S4 | S5 | S6 | S7 | S8 | S9 | S10 | Total |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Bacillariophyceae | 2,415 | 2,255 | 3,094 | 2,770 | 2,119 | 2,265 | 1,883 | 1,922 | 1,893 | 1,196 | 21,812 |
| Dinophyceae | 219 | 241 | 401 | 199 | 316 | 247 | 274 | 159 | 370 | 19 | 2,445 |
| Chlorophyceae | 703 | 1,271 | 863 | 833 | 696 | 620 | 632 | 499 | 566 | 428 | 7,111 |
| Cyanophyceae | 715 | 977 | 897 | 715 | 753 | 524 | 484 | 335 | 438 | 280 | 6,118 |
| Chrysophyceae | 0 | 4 | 15 | 10 | 8 | 6 | 14 | 9 | 2 | 3 | 71 |
| Dictyochophyceae | 1 | 2 | 6 | 2 | 2 | 4 | 1 | 3 | 1 | 0 | 22 |
| Rhodophyceae | 10 | 8 | 4 | 3 | 8 | 13 | 15 | 10 | 7 | 14 | 92 |
| Haptophyceae | 6 | 7 | 11 | 15 | 5 | 10 | 7 | 8 | 8 | 2 | 79 |
| Eustigmatophyceae | 8 | 57 | 25 | 27 | 32 | 8 | 3 | 16 | 4 | 1 | 181 |
| Cryptophyceae | 6 | 0 | 0 | 7 | 3 | 2 | 7 | 4 | 1 | 0 | 30 |
| Euglenophyceae | 37 | 25 | 61 | 72 | 54 | 43 | 36 | 76 | 106 | 1 | 511 |
| Total | 4,120 | 4,847 | 5,377 | 4,653 | 3,996 | 3,742 | 3,356 | 3,041 | 3,396 | 1,944 | 38,472 |

**TABLE A2** Correlation matrix of selected parameters and dominant phytoplankton classes in interconnected channels, main waterbody (bold) and reference site (italics)

| | BAC | DIN | CHL | CYA | BAC | DIN | CHL | CYA | BAC | DIN | CHL | CYA |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| AT | −0.187 | −0.234 | −0.113 | −0.653 | **−0.988** | **−0.478** | **−0.864** | **0.999*** | *0.986* | *−0.34* | *0.462* | *−0.383* |
| WT | −0.839 | −0.864 | −0.795 | −0.999* | **−0.846** | **−0.938** | **−0.982** | **0.783** | *0.893* | *0.282* | *−0.151* | *−0.855* |
| pH | −0.997* | −0.992 | −1.000** | −0.826 | **−0.997*** | **−0.549** | **−0.903** | **0.999*** | *0.018* | *0.98* | *−0.945* | *−0.858* |
| SL | 0.719 | 0.685 | 0.769 | 0.275 | **0.523** | **0.995** | **0.797** | **−0.426** | *0.501* | *−0.942* | *0.978* | *0.47* |
| FC | −0.993 | −0.987 | −0.999* | −0.803 | **−0.014** | **−0.801** | **−0.375** | **−0.096** | *−0.995* | *0.086* | *−0.218* | *0.608* |
| AL | −0.857 | −0.88 | −0.815 | −1.000* | **−0.66** | **−0.998*** | **−0.887** | **0.573** | *−0.445* | *−0.8* | *0.713* | *0.995* |
| BOD | −0.46 | −0.417 | −0.526 | 0.045 | **0.697** | **0.993** | **0.91** | **−0.614** | *−0.512* | *0.937* | *−0.975* | *−0.458* |
| PT | −0.731 | −0.697 | −0.78 | −0.292 | **0.298** | **−0.575** | **−0.069** | **−0.401** | *−0.782* | *−0.472* | *0.35* | *0.943* |
| NT | −0.768 | −0.798 | −0.718 | −0.985 | **−0.826** | **−0.057** | **−0.566** | **0.883** | *0.165* | *0.94* | *−0.886* | *−0.924* |
| AMN | −0.652 | −0.615 | −0.708 | −0.186 | **0.741** | **−0.08** | **0.448** | **−0.811** | *0.384* | *−0.978* | *0.997** | *0.581* |
| SLC | −0.781 | −0.826 | −0.365 | −0.365 | **−0.231** | **−0.912** | **−0.568** | **0.123** | *−0.949* | *−0.138* | *0.005* | *0.769* |

*Note:* (*5% level significant ($p < .05$; **1% level significant ($p < .01$)).

Abbreviations: ALK, alkalinity; AMN, ammonia–nitrogen; AT, atmospheric temperature; BAC, Bacillariophyceae; BOD, biological oxygen demand; CHL, Chlorophyceae; CTSI, Carlson trophic state index; CYA, Cyanophyceae; DIN, Dinophyceae; Fc, free carbon dioxide; NT, nitrate; PT, phosphate; SL, salinity; SLC, silicate; WT, water temperature.

**TABLE A3** Correlation between selected raw effluent characteristics with dominant phytoplankton and zooplankton classes in interconnected channels (black) and in main waterbody (italics)

| | pH | pH | BOD | BOD | NT | NT | AMM | AMM | BAC | BAC | DIN | DIN | CHL | CHL | CYA | CYA | TIN | TIN | COP | COP | CLA | CLA | ROT | ROT |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| pH | 1 | *1* | −0.464 | *−0.858* | −0.74 | *−0.106* | −0.963 | *−0.974* | −0.961 | *−0.04* | −0.973 | *0.208* | −0.938 | *0.402* | −0.97 | *−0.975* | 0.281 | *0.68* | 0.03 | *−0.97* | −1 | *0.313* | −0.999$^{*}$ | *0.043* |
| BOD | - | - | 1 | *1* | 0.939 | *−0.419* | 0.685 | *0.721* | 0.69 | *−0.48* | 0.655 | *−0.681* | 0.743 | *−0.815* | 0.237 | *0.724* | −0.98 | *−0.96* | 0.87 | *0.697* | 0.535 | *−0.76* | 0.428 | *−0.55* |
| NT | - | - | - | - | 1 | *1* | 0.893 | *0.327* | 0.897 | *0.998*$^{*}$ | 0.874 | *0.95* | 0.927 | *0.868* | 0.555 | *0.323* | −0.85 | *0.66* | 0.65 | *0.359* | 0.792 | *0.911* | 0.712 | *0.989* |
| AMM | - | - | - | - | - | - | 1 | *1* | 1.000$^{**}$ | *0.264* | 0.999$^{*}$ | *0.017* | 0.997 | *−0.185* | 0.87 | *1.000*$^{**}$ | −0.53 | *−0.5* | 0.24 | *0.999*$^{*}$ | 0.982 | *−0.09* | 0.951 | *0.182* |
| BAC | - | - | - | - | - | - | - | - | 1 | *1* | 0.999$^{*}$ | *0.969* | 0.997$^{*}$ | *0.899* | 0.866 | *0.26* | −0.54 | *0.7* | 0.25 | *0.297* | 0.981 | *0.937* | 0.949 | *0.996* |
| DIN | - | - | - | - | - | - | - | - | - | - | 1 | *1* | 0.992 | *0.979* | 0.889 | *0.013* | −0.49 | *0.86* | 0.2 | *0.051* | 0.989 | *0.994* | 0.963 | *0.986* |
| CHL | - | - | - | - | - | - | - | - | - | - | - | - | 1 | *1* | 0.826 | *−0.19* | −0.6 | *0.94* | 0.32 | *−0.15* | 0.963 | *0.995* | 0.923 | *0.932* |
| CYA | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | *1* | −0.04 | *−0.5* | −0.3 | *0.999*$^{*}$ | 0.947 | *−0.1* | 0.979 | *0.178* |
| TIN | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | *1* | −1 | *−0.47* | −0.36 | *0.908* | −0.242 | *0.76* |
| COP | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | *1* | 0.052 | *−0.06* | −0.071 | *0.216* |
| CLA | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | *1* | 0.992 | *0.963* |
| ROT | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | *1* |