UNIVERSITY OF KWAZULU-NATAL

The dynamics of nano- and microplankton in the St. Lucia estuarine lake system, KwaZulu-Natal.

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Dedication

I dedicate this research to my loving grandmother *MAMI FAMBO LYDIA KIEN*.

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Abstract

St. Lucia estuarine lake system has a history of episodic droughts and floods leading to a high variability in physico-chemical characteristics which may influence the community structure of nano- and microplankton that are the main primary energy synthesisers in aquatic systems. Originally, the St. Lucia estuary and the Mfolozi River shared the same mouth with the Mfolozi River acting as the main source of fresh water to the system and as stabiliser of the common inlet. Due to prolonged mouth closure from 2001-2012 and high evaporation rates, severe extended droughts and high salinities were experienced in 2001-2012.This project aimed to characterise community composition and biomass of nano-and microplankton (autotrophic and heterotrophic) under the varying and extreme physico-chemical conditions in order to characterize the energy basis of the planktonic food web in St. Lucia. Its main objectives were to compare and add onto Johnson's (1977) list of phytoplankton taxa in the system, to estimate the biomass (carbon) of nano- and microplankton from cell counts and biovolume measurements, and lastly to understand and establish trends in the change in community structure of these organisms with the varying physico-chemical characteristics.

Nano- and microplankton samples were collected monthly from October 2010 to September 2011 at three different sites: Lister's Point, Charters Creek and the mouth representing the lakes and estuary Channel. Chlorophyll a and physico-chemical parameters were also measured *in situ* during collection. In the lab, samples were settled using the Utermöhl method and species were identified to at least genus level, counted and cell measurements taken under an inverted microscope for biovolume calculations and biomass thereof. Abundance in cells per liter and biomass (carbon) in pg/L was then analyzed from the counts.

The nano-and microplankton groups recorded in the system were cyanobacteria, chlorophytes, cryptophytes, dinoflagellates, ciliates and diatoms. Seventy eight phytoplankton taxa were identified composed of 56 diatoms, eight green algae, one cryptophyte, seven cyanobacteria and six dinoflagellate taxa. Nineteen ciliate taxa were also found. Only 12 of the diatom taxa identified in this study were listed by Johnson (1977), none of the taxa in the other phytoplankton groups was listed by Johnson (1977). The Johnson (1977) study conducted in the system from 1975-1977 listed the phytoplankton taxa occurring at that time. There was no significant difference in the community composition, biovolume and biomass between seasons hence no seasonal trend however, there were significant differences in the nano- and microplankton community composition, biovolume and biomass at the three different sites of the system. Cyanobacteria were the main taxa in the northern embayments dominating in abundance, biovolume and biomass (biological variables), green algae and cryptophytes dominated in abundance, biovolume and

biomass in the Channels while in South Lake, green algae dominated in abundance but diatoms dominated in biovolume and biomass. Ciliate biological variables were higher in the northern regions than in the other parts of the estuary. The absence or limited grazing pressure of ciliate predators in the northern region due to their inability to cope with the extreme salinities compared to the other parts of the system explains why the northern embayments had the highest abundance, biovolume and biomass of ciliates. Ciliates and heterotrophic dinoflagellates were the heterotrophs in this study. Autotrophic:heterotrophic biomass ratio was lowest in the northern regions as heterotrophs had a higher biomass there. This ratio was higher in the South Lake and the Channel. The lack of stratification and generally high turbidity in the system made the system unfavourable for dinoflagellate growth. The higher presence of ciliate predators in the South Lake and Channel probably accounts for the low heterotrophic biomass hence high autotrophic:heterotrophic biomass ratio in South Lake and the Channel.

Nutrients were not limiting during this study and salinity was the main physico-chemical characteristic accounting for the differences in nano- and microplankton biological variables. The *Cyanothece* bloom in the northern region was primarily due to high salinities (>150) which also indicated unfavourable conditions for other plankton types. The high diatom biomass in the southern lake was due to low salinities (<28) which favoured their growth, whereas chlorophytes and cryptophytes dominated in the Channel mainly due to low turbidity (median of 11.4NTU) and fresh water input from the Mfolozi lowering salinities (<5).

The South Lake and Channel thus had the highest available energy for higher trophic level organism since 1) diatoms and green algae are the most favoured food source for phytoplankton grazers while the cyanobacteria though most abundant are the least favoured food source leaving the northern lake with smaller energy source for higher trophic level organisms and 2) The low autotrophic:heterotrophic biomass ratio in the northern region leaves the region with a lower net carbon biomass than the other parts of the system with a higher autotrophic: heterotrophic biomass.

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CHAPTER ONE

LITERATURE REVIEW

1.1 Estuaries and Estuarine lakes

An estuary is defined by Potter et al (2010) as "a partially enclosed coastal body of water that is either permanently or periodically open to the sea and which receives at least periodic discharge from a river(s), and thus, while its salinity is typically less than that of natural sea water and varies temporally and along its length, it can become hypersaline in regions when evaporative water loss is high and freshwater and tidal inputs are negligible". South Africa has approximately 289 estuaries classified into five main groups by Whitfield (1992) as estuarine bays, permanently open, river mouth, temporarily open estuaries and estuarine lakes. Seventy three of these estuaries are found in the KwaZulu-Natal (KZN) coastline only two of which are estuarine lake systems: St. Lucia estuarine system and Kosi Bay estuary (Begg, 1978).

Estuarine lakes are large bodies of water separated from the sea by vegetation sand dunes. Estuarine lakes also have rivers discharging into them and from time to time are connected to the sea however if they lose their connection with the sea, they get isolated and develop into coastal lakes (Whitfield, 1992). Generally in estuaries, the interaction between salt water from the sea and freshwater from rivers as a basis of estuarine hydrodynamics compounded by other factors such as channel structure, sediment movement, effects of wind, waves, isolation, anthropogenic inputs and biotic processes make estuaries a very dynamic ecosystem (Schumann & Largier, 1999). The hydrodynamics of estuarine lakes is determined by the depth of the system with wide and shallow systems being more influenced by wind (Schumann &Largier, 1999). Estuaries may become hypersaline in the upper reaches of tidal influx and or when fresh water inflow is limited. In shallow estuaries, surface water gravity and winds which forms surface waves better explain the water movements on a short time scale. This water movement causes suspension of benthic plankton and also prevents vertical salinity gradients to build up. In KZN during summer, estuaries are influenced by easterly winds associated with the Indian Ocean while in winter, the westerly winds prevail (Schumann & Largier, 1999).

1.1.1 Importance of estuaries

Estuaries receive nutrient rich waters from inflowing rivers. This makes estuaries to have higher nutrient concentrations compared to its surrounding aquatic systems hence the potential for primary production is high. This food coupled with high turbidity which acts as protection of juvenile fish and other planktonic organisms from predators and the relatively stable environment due to less tidal influence, makes

estuaries an important breeding and nursery ground for various marine organisms. These organisms include a number of seasonal marine fish of high commercial and conservation value (Whitfield, 1977; Perissinotto et al., 2003) and prawns (Forbes & Demetriades, 2005). Some of these fish such as *Argyrosomus thorpei* (Square tail kob), *Caranx ignobilis* (Giant kingfish) and *Rhabdosargus sarba* (Natal stump nose) are of economic value, hence food and source of income to local inhabitants of the estuarine community.

Estuaries have also been known for their rich bird life. Whitfield and Blaber (1978) recorded 340 bird species 90 which were dependent on the St. Lucia estuarine lake system. Estuaries in tropical and subtropical regions are well known as a sanctuary for larger reptiles and mammals such as crocodiles and hippopotamus respectively. Estuaries are not only a hotspot for diversity, but also a source of recreation and income for the area and country at large as they attract tourists who come to enjoy its scenario, wild life, birds Hocky & Turpie, 1999), sport fishing and other general water sports as well (Adams et al., 2011)

1.2 Nano- and microplankton

Plankton can be defined as partially drifting or wandering forms of protists which live freely in waters and are at the mercy of waters movements for their movements. Many of these organisms are immobile while others have a limited ability to swim and thus change position in the water column. Plankton are highly diverse in size, form, pigmentation and growth rate (Haris, 1986; Mitra, 2004). Plankton are divided into groups according to size with size ranging from less than 5 µm to several centimeters: 1mm- several centimeters are animals, 500µm- 1mm are mainly animals and few phytoplankton, 60µm to 500µm are mostly phytoplankton (mainly peridians, diatoms), ciliates, few eggs, and larval forms, 5µm – 60µm are made up of small diatoms, flagellates, ciliates and lastly, less than 5µm are mainly small flagellates and bacteria (Hoppenrath et al., 2009). Some plankton however are up to a few meters in size e.g. some jellyfish (Suthers & Rissik 2009). Nano- and microplankton fall within the size range of 2 – 200 µm and could either be photosynthetic, heterotrophic or mixotrophic. These organisms represent important components of the plankton in all aquatic ecosystems. The carbon biomass of these planktonic organisms is an important parameter in the construction of ecosystem models and biogeochemical carbon budgets (Menden-Deuer & Lessard, 2000). The carbon content of some major planktonic groups can be used for ecosystem models and to analyse for temporal and spatial variability in the primary production of a system (Menden-Deuer & Lessard, 2000). Estuaries are very dynamic ecosystems with high spatio-temporal variability of planktonic community structure (Azémar et al., 2007). For this study, nano- and microplankton will be split into 2 groups: phytoplankton and microzooplankton.

1.2.1 Phytoplankton

Also called plant plankton, phytoplankton is the group of plankton that uses energy from the sun, carbon dioxide, dissolved inorganic nutrients and salts to produce organic matter in aquatic ecosystems (Reynolds, 2006). They are found in the water column with their spatial distribution, horizontal and vertical position being controlled mainly by water motion (Haris, 1986). However, phytoplankton also tends to concentrate around areas with high nutrients and or light in the water column. Phytoplankton can either be prokaryotic or eukaryotic with sizes ranging from as small as bacteria (<2 µm) to large sizes (up to 2000µm) such that some dinoflagellates are visible with the naked eye. Phytoplankton size fractions are: <2 µm are picophytoplankton, 2-20 µm are nanophytoplankton, 20-200 µm are microphytoplankton and 200- <2000 µm are macrophytoplankton (Finkel et al., 2010).Their growth rates vary from a few doublings per day to one doubling in every week or ten days (Haris, 1986) and their life span varies on a time scale of hours to a few days (Marra, 2002). The most common phytoplankton groups are the bacillariophytes, chrysophytes, chlorophytes, crytophytes, dinophytes,cyanophytes and xanthophytes (Haris, 1986). Phytoplankton in estuarine systems is dominated by bacillariophytes (diatoms) (Johnson, 1977; Huang, 2004; Dijkman & Kromkamp, 2006; Ramdani et al., 2009). According to Dijkman & Kromkamp (2006), the phytoplankton of the Scheldt estuary situated in the border region of Belgium and Netherlands is dominated by diatoms with a minor contribution from chlorophytes, cryptophyes, dinophytes and haptophytes. The Pearl River estuary of China also records diatoms as the most abundant group (Huang, 2004). In North African Coastal lagoons of MerjaZerga in Morocco and Ghar El Mehl in Tunisia, phytoplankton taxa include Dinophyceae, Bacillariophyceae, Cyanophyceae, Chlorophyceae, Euglenophyceae, Chrysophyceae, Cryptophyceae and Silicophyceae (Ramdani et al., 2009). This composition is the same for brackish lakes of Egypt, e.g. Lake Manzala(Ramdani et al., 2009), and South African estuaries such as Berg, Palmiet, Goukou, Gourits, Great Bank, Keurbooms, Gamtoos and Sundays with the exception of Chrysophyceae, Cryptophyceae and Silicophyceae that have not been recorded (Adams & Bate, 1999). In calm waters, phytoplankton could settle and be collected as benthic microflora, likewise, during disturbance and turbulence especially in shallow waters benthic microflora might be suspended in the water column and collected as phytoplankton (Wimpenny, 1966; Allanson& Baird, 1999).

1.2.1.1 Importance of phytoplankton in estuaries

Nano- and microphytoplankton are photosynthetic and the main primary synthesizers of organic matter (carbon) in aquatic ecosystems forming the bases of the trophic relationships in the ecosystem (Fielding et al., 1991; Ramdani et al., 2009; Konoplya & Soares, 2011). Phytoplankton is thus the most important group of organisms in an aquatic system as they determine the carbon stock available at the base of the food web (Mitra, 2004). They are a good food source for other protists, invertebrates and fish (Azémar et

al., 2007; Urrutxurtu, 2003; Grinienė et al., 2011). Nano- and microphytoplankton biomass and composition is thus used to predict fisheries potential (Hallegraeff et al., 2010) as phytoplankton are an energy source for invertebrates and larva which are an energy source for fish and their fries. Phytoplankton community structure changes with varying environmental conditions and hence can be used as an indicator of the ecological condition of a system (O'Boyle and Silke, 2010; Ramdani et al., 2009). Some species are good indicators of either high or low water quality (Lemoalle, 1998; Ramdani et al., 2009). Water quality is also defined by the chlorophyll a (chl a) content, poor is = >20 µg/L, fair = 5-20 µg/L and good = 0-5 µg/L (Borja & Basset, 2012). Diatoms flourish in good water quality while cyanobacteria and dinoflagellates are mostly recorded to dominate in poor water quality (Lec et al., 2008). Not only can phytoplankton monitoring be used to predict water quality, it can also be used as a warning sign for potential harmful algal blooms. Although harmful algal blooms are not a common occurrence in South African estuaries, algal blooms have been recorded in some South African estuaries such as the Sundays estuary (Kotsedi, 2011), St. Lucia estuarine lake (Muir & Perissinotto, 2011) and the Gamtoos estuary (Snow, 2000).

1.2.1.2 Phytoplankton diversity and community structure in estuaries

There exist spatio-temporal variations in phytoplankton diversity and community structure in all aquatic systems. In South Africa, most studies on estuarine phytoplankton have concentrated on total chl a, size fractionated chl a and very few on productivity. The use of chl a has limitations as it does not distinguish between the different groups or species (Caseet al., 2008) and does not give actual community composition of phytoplankton in the systems. It is important to know the actual species composition as phytoplankton undergo continual changes in its community composition and structure as grazers have different preferences for phytoplankton groups. Factors affecting the community structure (taxa composition, abundance, biovolume and biomass) of phytoplankton in estuaries include nutrients, light, turbidity, temperature, salinity (Haris, 1986; Reynolds, 2006; O'Boyle & Silke, 2010) grazing (Day et al., 2013) and the hydrology of the system (Schumann & Largier, 1999). In South African estuaries, phytoplankton diversity, growth and biomass are mainly influenced by nutrients and salinity (Adams & Bate, 1999).

Increase in phytoplankton biomass is said to be linked to the availability of nutrients (Reynolds, 2006). The availability of nutrients in an estuarine system depends greatly but not exclusively on water inflow from rivers. Freshwater brings in nutrients, causes the mixing of these new nutrients and already available nutrients in the estuarine sediment making more nutrients available to phytoplankton and other protozoans. However, in cases where freshwater inflow from rivers is high (floods) and has little resident time for the plankton to assimilate the nutrients, all nutrients are washed off to the sea and phytoplankton

biomass reduces as was observed in the Palmiet estuary (Largier & Taljaard, 1991). Chlorophyll a values in South African estuaries of Great Brak and Keurbooms have been reported to increase from 1 µg l^{-1} to 13 µg l^{-1} due to nutrients brought in by the river (Adams & Bate, 1999). A defined succession pattern of phytoplankton communities based on nutrient availability in estuaries which happen to be supported by studies on South African estuaries is that flagellates dominate when nutrients are depleting and stratification sets in while diatoms dominate when the nutrients are available and the system is well mixed (Adams & Bate, 1999). Dinoflagellates are said to dominate when the system is stable, stratified and the nutrient concentrations are high (Margalef, 1978; Felip & Catalan, 2000) as was reported for the Sundays estuary by Hilmer & Bate (1991). Other sources of estuarine nutrients include benthic regenerated nutrients and anthropogenic sources e.g. agricultural fertilizer flow as in the Sunday estuary (Adams & Bate, 1999). Nitrogen (N), Phosphorus (P) and Silica (Si) are the major nutrient elements phytoplankton requires with a host of other elements known as trace elements (Reynolds, 2006). Given the fluctuations of physico-chemical parameters in an estuary, the Redfield ratio of 106:16:1 for C:N:P (Redfield, 1934) is not always the optimal ratio for all species(Reynolds, 2006). Different species need different nutrients in different proportions however, the availability of N, P, Si and other trace elements such as Iron (Fe) in the system determines which phytoplankton group flourishes at a particular time. This is evident as a decrease in nitrate and ammonia concentrations in a system leads to the replacement of diatoms by nitrogen fixing cyanobacteria cells (Tyrell, 1999; Finkel et al., 2010). Silicon is also a limiting factor for diatoms since Si is used in the formation of diatom frustules. High amounts of nutrients especially P in an estuarine system often lead to blooms and eutrophication. In eutrophic systems in South Africa where nutrients are not limiting, salinity is the factor that determines productivity and growth in the phytoplankton community (Adams & Bate, 1999).

Estuarine systems with a greater horizontal salinity gradient have a higher phytoplankton and protozoan diversity than estuaries with a greater vertical salinity gradient (Adams & Bate, 1999). Increases in freshwater supply increase the horizontal salinity gradient of the system. Different phytoplankton and protozoan species are adapted to different salinity regimes, thus with an increase in the salinity gradient, a greater diversity of plankton is accommodated (Allanson& Baird, 1999). Certain diatoms occur across a wide range of salinities from marine conditions (salinity 35) to freshwater conditions (salinity <4) however others are adapted only to freshwater or are exclusively marine in nature and others brackish (salinity of 5-20) (Reynolds, 2006). Diatoms are euryhaline and are present throughout the estuarine system at all periods (Urrutxurtu, 2003) but high concentrations are reached by freshwater species (Lucas, 1986; Urrutxurtu, 2003). In South African estuaries, salinity has a seasonal pattern. Lowest values are recorded in spring and early summer while highest values are recorded in winter (Froneman, 2002).

Although little is recorded about the abundance of different phytoplankton species in South African estuaries, phytoplankton succession has not been recorded with respect to seasons. However, in estuaries of the temperate regions, dinoflagellates have a high abundance in late summer while green algae are more common in freshwater systems with highest abundances in late spring (Urrutxurtu, 2003; Huang, 2004; Ramdani et al., 2009).

A variation in pH can also affect the phytoplankton growth by changing the buffering system of the estuarine bed which thus influences the availability of trace elements and essential nutrients from the estuarine bed. In fresh water systems, a significant change in community structure of phytoplankton has not been recorded with fluctuations in pH (Chakraborty et al., 2011) however, chl a concentrations increase gradually as the system changes from acidic to neutral pH recording maximum chl a concentrations at a pH of 8.5. Green algae have a higher tolerance for a vast range of pH (Chakraborty et al., 2011). Most major ions in the adjacent sea waters and running rivers are not required as a main nutrient source by phytoplankton, though blue-green algae e.g. *Spirulina* was shown to be dominant in African lakes with high alkalinity (Haris, 1986). Diatoms, green algae and cyanobacteria assemblages have also been shown to be linked with the composition of ions (Haris, 1986).

Phytoplankton being photosynthetic in nature needs amongst other components, solar energy in the form of light for photosynthesis. The amount of light available for the cells determine to a large extent the primary production capacity of the cells. Over exposure to light might lead to photo-inhibition thus reduced primary production. The time spent by a cell in the euphotic zone and the degree of exposure to light is highly dependent on turbidity in estuarine systems as particulate matter in turbid waters interferes with the light penetrating the estuary. Turbidity which is a measure of the amount of particulate matter in the water column is influenced by water inflow, salinity, sediment grain size, wind, waves and tides. Water inflow from rivers causes turbulence which re-suspends already existing particulate matter and also brings in new sediment and particulate matter, thus increasing turbidity. However the rate of sediment re-suspension depends on its grain size as sediments with smaller grain size will be easily re-suspended. The increase in salinity gradient induces horizontal stratification that reduces turbidity (Froneman, 2002). Wave action during open mouth stages of an estuary and wind are important sources of turbulence especially in cases of shallow, wide estuarine lakes to influence increase turbidity. Larger organisms such as hippopotami and crocodiles whose presence causes re-suspension of particulates in estuarine systems also influence turbidity in estuarine systems. Turbidity is thus said to be one of the main factor determining phytoplankton productivity and chl a concentrations. This is true for most large and deeper

estuaries but is not the case with South African estuaries as they are generally shallow (<0.9m) and the light rays reach the estuarine bottom (Grange & Allanson, 1995; Adams & Bate, 1999).

It is important to consider the effect of zooplankton grazing on the diversity and community structure of phytoplankton. Some of the species e.g. diatoms are more desirable hence easily grazed upon while others like larger dinoflagellates and cyanobacteria are immune to grazing or less grazed upon by zooplankton (Moss & Booker, 1989) as dinoflagellates and cyanobacteria could produce toxins or chemicals to keep the grazers away. In some TOCEs, zooplankton can graze up to 70% of the available phytoplankton biomass (Kibirige & Perissinotto, 2003).

1.2.1.3 Phytoplankton productivity

Phytoplankton productivity, i.e. primary production, contributes immensely to the total production in shallow intertidal waters (Krompkanp, 1991). Primary production is the measure of the amount of organic carbon (C) assimilated by photosynthetic organisms (Geider & MacIntyre, 2002). In order to get a complete understanding of primary productivity by phytoplankton, biomass measurements, yield and rate of transfer of C to higher trophic level organisms should be measured (Marra, 2002). Algal biomass and cell numbers are a measure of successful conversion of inorganic C to organic C (Gosselain, 2000), but disregard grazing. Most often, chl a values are used as estimates of phytoplankton biomass and can be converted to carbon biomass by using a C:chl a ratio (Gosselain, 2000). However, when phytoplankton biomass is measured through cell counts and biovolume measurements, biomass as carbon is estimated by using linear equations of cell dimensions suggested for each algae group (Menden-Deuer & Lessard, 2000).

Light, temperature, and nutrient limitations affect the C:biovolume ratio (Thompson, 1992;Montagnes et al., 1994; Felip & Catalan, 2000;Geider & MacIntyre, 2002). Light and temperature have a positive correlation with cell carbon. Nutrients (phosphorus and nitrogen) also have a positive correlation with chl a and carbon (Geider & MacIntyre, 2002) depending on N:P ratios. The source of nitrogen in particular matters as nitrates increase the chl a:RUBISCO (a catalytic enzyme in the Calvin cycle of Photosynthesis, accounting for 1-10% of cell carbon and accounts for up to 5 times as much cell mass as chl a under high light conditions) ratio when compared to ammonia (Geider & MacIntyre, 2002). In estuaries with a regular inflow of freshwater, such as in the Gourits estuary in South Africa, high chl a values (>20µg L^{-1}) are recorded due to high nitrate concentrations. This was the same for the Sundays Estuary where values up to 29 µg L^{-1} were recorded (Adams & Bate, 1999). Generally in South African estuaries, average

phytoplankton biomass as chl a values range from as low as 0.01 μg L^{-1} to 24.08 μg L^{-1} (Perissinotto et al., 2003).

Species composition and cell size influence the C:chl a ratio (Felip & Catalan, 2000; Gosselain, 2000; Geider & MacIntyre, 2002). Chlorophyll a content is inversely related to biovolume per unit cell (Felip & Catalan 2000) hence smaller cells (<20 μm) contain a relatively higher amount of chl a compared to larger (>20μm) cells. Compared to other phytoplankton cells of the same size, diatoms have the least amount of carbon per unit cell volume due to the large vacuole they possess (Gosselain, 2000; Menden-Deuer & Lessard, 2000; Menden-deuer et al., 2001). Green algae have a higher chl a content per unit volume than dinoflagellates (Felip & Catalan, 2000). However, phytoplankton species with a biovolume of <3000 μm^3 have similar C:biovolume ratios (Montagnes et al., 1994; Menden-Deuer & Lessard, 2000). There is a good agreement between measured biomass from chl a values and biomass from biovolume measurements of phytoplankton cells (Gosselain, 2000).

Very few phytoplankton studies in South Africa have investigated the primary production of phytoplankton and they suggest that estuaries are very productive ecosystems (Adams & Bate, 1999). Some examples of primary production rates in South African estuaries include: 900 mg C m^{-2} per day for Sundays estuary (Hilmer, 1990), 9.083-10.500 mg C m^{-2} h^{-1} for St. Lucia estuarine system (Fielding et al.,1991; van der Molen & Perissinotto, 2011), 51-216 mg C m^{-2} h^{-1} for the Mpenjati estuary and 1-340 mg C m^{-2} h^{-1} for the Mdloti estuary (Anandraj, 2007). These values are within range and higher than values (0.811-62.790 mg C m^{-2} h^{-1}) recorded in European and North American estuaries (Tillman et al., 2000, Struski & Bacher, 2006).

1.2.2 Microzooplankton

1.2.2.1 Ciliates

Apart from phytoplankton, there are other protist and plankton in aquatic ecosystems. These other protists some of which include the groups Apicomplexa, Foramifera, Amoebozoa and Ciliophora could be heterotrophic or mixotrophic as is the case with ciliates. Apart from phytoplankton, ciliates dominate microzooplankton abundance in some South African estuarine ecosystems e.g. in the Mpenjati (Kibirige et al., 2002) and Kasouga estuaries (Wasserman et al., 2013). Ciliates are a major component of pelagic food webs in aquatic ecosystems. They do not only constitute part of the microbial food web (Urrutxurtu, 2003; Grinienė et al., 2011) but also constitute crucial parts of the herbivorous web as they feed on a wide range of organisms which include bacteria, phytoplankton and other ciliates (Urrutxurtu, 2003; Grinienė et al., 2011). Ciliates are said to clearly dominate the grazing pressure on phytoplankton in some

freshwater lakes in seasons when they are abundant and are a good food source for meso- and microzooplankton with its main predators being rotifers, cladocerans, copepods (Urrutxurtu, 2003; Azémar et al., 2007) and other higher trophic level organisms. Thus ciliates constitute an energy link between low trophic level organisms and higher trophic level organism (Pfister, 2002; Grinienė et al, 2011). Ciliates have very high metabolic rates, short generation time (Pfister, 2002; Grinienė et al., 2011), rapid growth and high turnover rates (Pfister, 2002; Du et al., 2012). These characteristics allow ciliates to respond rapidly to changing environmental factors hence some species are used as bioindicators in water quality analysis (Pfister, 2002). Also these characteristics enable ciliates to play an important role in determining the overall grazing rates, nutrient generation and secondary production in estuarine systems, especially when they are abundant (Urrutxurtu, 2003; Grinienė et al., 2011).

Very little is recorded about ciliates in South African estuaries however, in European estuaries, ciliates have been suggested to be cosmopolitan in nature (Urrutxurtu, 2003) but their community structure varies with time and space(Al-rasheid & Sleigh 1995; Urrutxurtu et al., 2003; Grinienė et al., 2011). Amongst other factors, salinity (Pfister, 2002; Du et al., 2012), temperature and food availability (phytoplankton, bacteria and other ciliates) determine the type of ciliate species and their abundance in a particular region, at a particular time (Urrutxurtu, 2003; Grinienė et al., 2011). In the temperate regions, highest abundance and diversity of ciliates occurs in late spring to summer and lowest abundance and diversity occurs in autumn to early spring (Pfister, 2002; Urrutxurtu, 2003;Grinienė et al., 2011). An example is the Nervion river estuary in Spain (43°20'N, 3°00' W) with counts of 7-5.4x10^5 Cells L^{-1}in late spring to summer and 100 Cells L^{-1} in autumn – early spring (Urrutxurtu, 2003). Ciliate abundance and diversity correlates positively with temperature and chl a but correlates negatively with salinity. In late spring to summer, temperatures are high, chl a values are also highest corresponding to the high availability of phytoplankton being the main source of food for ciliates. With the onset of autumn, temperatures fall and chl a values decline (Pfister, 2002; Urrutxurtu, 2003; Grinienė et al., 2011).

Along the estuary, the highest diversity and abundance of ciliates occurs in the most brackish regions as there is a mixture of marine and freshwaters ciliate species with freshwater species more readily adapting to marine waters than marine species to freshwater (Pfister, 2002). Also, brackish waters have a higher diversity and abundance of ciliates compared to fresh water lakes (Pfister, 2002). The least diverse and least abundant ciliate community is found towards high salinities of >24 as few species such as *Euplotes* sp. are adapted to osmotic stress (Pfister, 2002; Urrutxurtu, 2003). Different sites in an estuarine system may have unique species defined by environmental conditions (Pfister, 2002). The main orders found in an estuarine system are Heterotrichida, Choreotrichida, Cryptophorida, Oligotrichida, Protomastida,

Haptorida, and Pleurostomatida (Urrutxurtu, 2003; Grinienė et al., 2011). Some taxa associated with freshwater most likely to be found in the low salinity regions of estuaries are *Paramecium*, *Blepharisma*, some *Strombidium* (Al-rasheid & Sleigh, 1995) and members of the order Scuticociliatida (Urrutxurtu, 2003). Some taxa associated with brackish water include *Euplotes*, *Strombidium*, *Uronema*, *Didinium*, *Monodinium*, *Mesodinium*, *Pleuronema*, *Tintinopsis* and *Codonella* (Al-rasheid &Sleigh, 1995; Pfister, 2002). The marine, outer region would likely include the large Tintiniids and Haptorids (Urrutxurtu, 2003). The fresh, brackish and marine taxa listed above are cosmopolitan in nature.

1.3 Study Site

1.3.1 Origin

The St. Lucia estuarine lake system was formed by marine erosion during the early Holocene (Orme, 1973). Sea levels in this era were about 120 times lower than current sea values and the eroded depressions were river systems (Hill, 1975; Johnson, 1977). Six thousand years ago, sea levels increased covering the depressions including the present-day Mfolozi swamps and surrounding flood plains (Whitfield & Taylor, 2009). During the Flandrian transgression (18.000-19000 years ago when there was a rise in sea levels affecting coastal regions), the system was approximately 40 m deep but during the late Pleistocene (11700 years ago) and as the Holocene (8000 years ago) progressed, it accumulated sediments leading to its present shallow state with compartments less than 1 m in depth (Van Heerden, 1976).

1.3.2 Topography and History

The St. Lucia estuarine lake system (between 27° 52′S, 28° 24′S and 32° 21′E, 32° 34′E) situated in the northern parts of KwaZulu-Natal province of South Africa is the largest estuarine system in southern Africa and one of the largest estuarine lake systems in Africa (Day, 1981; Fielding et al., 1991; Whitfield & Taylor, 2009; Perissinotto et al., 2010). This system is part of the iSimangaliso Wetland Park which was previously called Greater St. Lucia Wetland Park and gained World Heritage Site status in 1999 (Taylor, 2006) after being declared a RAMSAR site in 1991(Taylor, 1991). The area covered by the system varies from 300-360 Km² depending on water levels (Begg, 1978).

The St. Lucia estuarine system is divided into three compartments: the northern embayments (which comprise False Bay and North Lake), the South Lake and the Channel (Figure 1). It has three interconnected shallow lakes (False Bay, North Lake and South Lake) with maximum depths of 3.5m recorded but an average depth of 0.9m (Perissinotto et al., 2010; Cyrus et al., 2011). These interconnected lakes discharge into the Channel, a 21 Km long Channel which empties to the Indian Ocean through the mouth located 1.5Km north of the Mfolozi River mouth (Orme, 1973;Day, 1981;Whitfield et al. 2009;

Perissinotto et al. 2010). The South Lake has tidal influence from the sea during the open mouth conditions while the northern embayments have very little or no tidal influence from the sea.

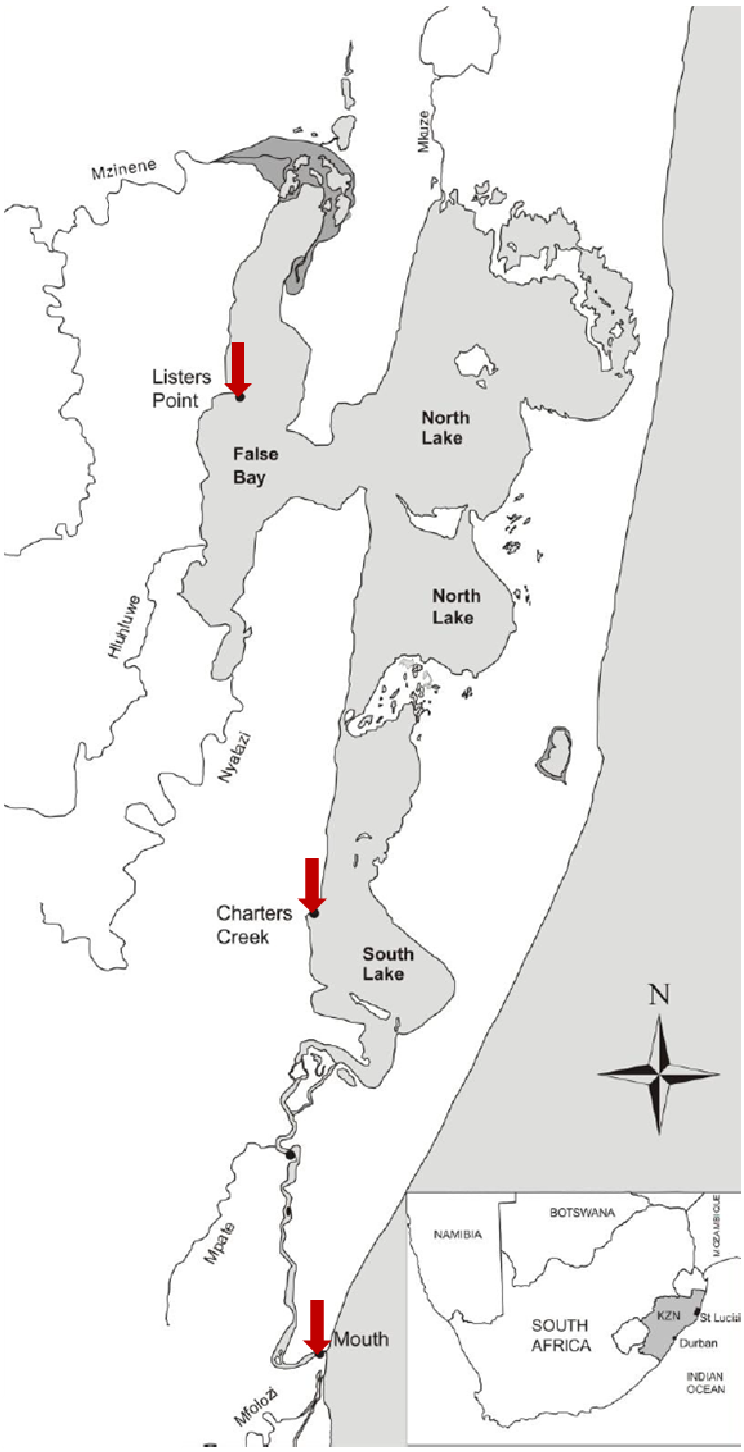

Figure 1: Map of Lake St Lucia with the sampling sites indicated by bold arrows. Provided by Sarah Bownes, created from data captured by the U.S. Geological Survey, Center for Earth Resources Observation and Science (EROS) (http://edcsns17.cr.usgs.gov/NewEarthExplorer).

The St. Lucia estuarine lake system is periodically open to the sea and has freshwater inputs from rivers and ground seepage (Perissinotto et al., 2010). The system normally experiences alternating dry and wet seasons due to climatic cycles leading to a high variability in its physico-chemical characteristics both temporarily and spatially. During the wet seasons, there is maximum input of freshwater and during the dry seasons, water from the Mfolozi River would flow up the Channel into the lakes keeping the water level frequently above sea level (Whitfield & Taylor, 2009).

In the early 1900's the system and the Mfolzi swamps began to be modified by human activities. The most prominent was the onset of sugar cane cultivation in 1914 in the delta area of the Mfolozi flats (Cyrus et al., 2010). In 1936, a canal called the Warner's Drain that was created along the Mfolozi River with the aim of removing flood waters from the floodplains to the sea which left the St. Lucia system void of its main source of freshwater during the dry seasons. It should be noted that the Mfolozi River not only acted as a source of freshwater, but its flood plains were a sediment filter such that only relatively sediment free water entered the St. Lucia estuarine system. After the canalization in 1936, the Mfolozi River which previously carried away sediments (estimated at 0.68×10^6 tons per year) (Whitfield & Taylor, 2009)from the estuarine mouth could no longer function in this capacity causing the sedimentation in the mouth of the estuarine bay to be permanent. The rate of sedimentation at the estuarine mouth increased such that during the droughts of the 1950's the common mouth area of the Mfolozi and St. Lucia estuarine system was blocked severing the connection of these 2 systems and the sea (Whitfield & Taylor, 2009). With more emphasis placed on the well being of the farms in the floodplains than the estuarine system, a separate link from the Mfolozi River to the sea was excavated 1.5 Km away from the current St. Lucia mouth (Kriel, 1966; Whitfield & Taylor, 2009). Though the newly created Mfolozi mouth showed a potential of reconnecting with the St. Lucia estuary mouth by migrating 60m per month towards the St. Lucia mouth, the Mfolozi mouth was kept apart by breaching it near Maphelane before it would reconnect (Whitfield & Taylor, 2009).

Prior to human modification of the system, the St. Lucia estuarine mouth would only close for a maximum of two years (Whitfield & Taylor, 2009). This was due to a shared inlet with the Mfolozi, which allowed for a predominantly open mouth due to its high runoff estimated at 920×10^6 m^3 per average year The St. Lucia mouth closed in 1950 and could not be naturally opened by floods from the river any more due to the separation of the common St. Lucia and the Mfolozi Mouth which caused the high sedimentation at the estuarine mouth (Taylor, 2006).

The mouth and riverine Channel was dredged and "Permanent" hard structures were set up to keep the mouth open. This led to phytoplankton of marine origin dominating the system (Johnson, 1977). Not only were these structures expensive to set up and maintain, they were washed up and destroyed by the Cyclone Domoina of 1984 and since then these structures were never replaced (Whitfield et al., 2009).

In 2002, the mouth closed with the onset of a drought in 2001 but was never opened artificially again. The mouth remained closed for 2 years and only opened briefly in 2004 when the Mfolozi River overtopped its banks flooding the system as well as during the Cyclone Gamede in March 2007 where the mouth stayed open for 175 days (Pillay & Perissinotto, 2009; Whitfield et al., 2009). Since August 2007 beyond the end of this study, the mouth has been closed.

1.3.3 Physico-chemical parameters

1.3.3.1 Freshwater resources and salinity

The St. Lucia estuarine system has a catchment area of 9000km²(Day, 1981) and receives freshwater supply from five rivers: Mpate, Mkuze, Mzinene, Hluhluwe and Nyalazi; direct rainfall, ground seepage and runoffs from the sand dunes between the eastern shores and the Indian ocean (Perissinotto et al., 2010) giving a total mean annual freshwater input of $563x10^6 m^3$ (Hutchison, 1976). Direct rainfall is said to provide 50% of the freshwater input (Taylor, 2006)while river runoff provides 46% and ground seepage provides the remaining 4% (Cyrus et al., 2010).This freshwater input leaves the system with salinities less than that of natural sea water and varying along its length. During this study, parts of the St. Lucia estuarine lake system was hypersaline with a reverse salinity gradient due to negligible freshwater input due to below average rainfall, high evaporation rates, and the closure of the mouth since 2007 (Whitfield et al., 2009).

The decline in annual rainfall in the KwaZulu-Natal province is a main reason for the decrease in freshwater supply to the system (Cyrus et al., 2010). This is evident by the steep downward trend of the cumulative deviation of monthly rainfall from the monthly mean rainfall recorded in December 1999-September 2009. It is estimated that during this period almost 1.2 m of precipitation that was expected to have entered the St. Lucia estuarine lake system did not occur due to the decrease in rainfall (Cyrus et al., 2010).

Agro-economic activities: forestry plantations, sugar cane farms and others around the system's floodplains have also greatly reduced the freshwater supply to the system. These agro-economic activities occupy very vast extents of land and have a high freshwater requirement. The tree plantations occupy

30700 ha, irrigated fields occupy 9300 ha, and farm dams occupy approximately 745 ha (Whitfield et al., 2009). The agro-economic constraints coupled with the fact that during the droughts (e.g. 1967-1972 drought) the rivers no longer provided freshwater, the already freshwater stressed system became even more stressed (Whitfield et al., 2009). Due to the fact that the system was already stressed and salts (20 million tons) (Taylor, 2006) were brought into the system by the artificial opening of the mouth, Hutchison (1976) proposed an addition of freshwater into the system in a bid to solve the unhealthy hypersaline state of the system. Hence, McGill (1980) proposed the re-linking of the Mfolozi and the St. Lucia estuarine lake system as the main way of solving this constraint. The importance of the Mfolozi River cannot be overemphasized as prior to the closure of Mfolozi link with St. Lucia, the annual freshwater input from the Mfolozi alone was $920x10^6$ m^3 and those of the other five rivers combined was only $362x10^6$ m^3. Moreover the most recent drought of 2000 till after this study saw all the other five river sources of freshwater dry out (Whitfield & Taylor, 2009).

By May 2003, the northern embayment was hypersaline and had lost all of its surface water by July 2006. It is estimated that by July 2006, the system had lost approximately 90% of its surface water (Whitfield & Taylor, 2009). To ameliorate this situation, a link was created between the Mfolozi and the St. Lucia estuarine lake system. However, up to the end of this study (2011) only the mouth, Channel and the South Lake received this freshwater. This link brought salinities in the Channel to <10 still leaving the Northern embayment hypersaline but providing water equivalent to 30mm of rainfall over the large 35000ha of the St. Lucia estuarine system (Whitfield & Taylor, 2009). Also in 2007, the pine plantations of the west and eastern shores where removed. This might increase the freshwater available to the system through ground water seepage (Whitfield & Taylor, 2009).

1.3.3.2 Salinity fluctuations

The main determinant of salinity in the St. Lucia estuarine system is the amount of freshwater input compared to the evaporation rate. This thus leads to seasonal variations in salinity based on the amount of rainfall with the winter months (dry season) generally having higher salinities than the summer months (rainy season).This is so because, in winter, the rate of evaporation is higher than the rate of freshwater input. The summer months have lower salinities since their high evaporation rates of approximately 75% of the total inflow is over shadowed by the higher freshwater input (Johnson, 1977; Day, 1981). During summer months, salinities generally are below 35 (brackish).Winter onsets with less rainfall causes the system, mostly the northern embayments to be hypersaline (>35) giving the system a reverse salinity gradient. In the absence of early summer rains, the whole system becomes hypersaline with salinities of >200 being recorded in 2003/2004 (Cyrus et al., 2010). These same reasons are evident in the salinity

fluctuations in the past decade (2000-2010) (Whitfield & Taylor, 2009). When the estuarine system is connected to the sea either during an open mouth phase or a cyclone event, the salinities in the estuarine system are approximately 35 (marine) (Cyrus et al., 2011; van der Molen & Perissinotto, 2011).

1.3.3.3 Turbidity and Irradiance

The St. Lucia estuarine lake system is a very turbid system with turbidity as high 250 NTU recorded before this study (Fielding et al., 1991) and being mainly influenced by the wind velocity (Johnson, 1977; Fielding et al., 1991). The shallow nature of the system coupled with the fine silts the rivers bring into the system, enables the wind to easily re-suspend settled material. According to Day (1954) wind velocity changes seasonally hence turbidity is expected to change seasonally too. Although this was not the case in measurements taken from 1973-1977 (Johnson, 1977), it was the case in 1987-1988, as turbidity was high in February coinciding with high wind velocity but low in July and September due to lower wind velocities (Fielding et al., 1991).

The amount of light measured as irradiance [Photosynthetic Available Radiation (PAR)] reaching cells is influenced by depth and turbidity (Muir & Perissinotto, 2011). Surface PAR is always higher than bottom PAR as light rays are deflected by particles as they move through the water column, e.g. surface PAR measured from 2004-2007 varied between 359-2250 μmol m^{-2} s^{-1} while the bottom recorded values ranging from 0-1596 μmol m^{-2} s^{-1}(Perissinotto et al., 2010). With respect to depth, the deeper the system, the less PAR reaching the bottom, e.g. the Mouth which is deeper recorded an average value of 81.8±2.5%irradiance,South Lake of intermediate depth; 70.2± 2.9%irradiance and the most shallow region, the North Lake had a mean value of 59.6±4.5%irradiance (Perissinotto et al., 2010).

1.3.3.4 Temperature

The temperature of the water column of the St. Lucia estuarine lake system is seasonal with variations depending on the depth of the system (Johnson, 1977; Perissinotto et al., 2010). The lowest mean temperatures e.g. 16-18°C (Day, 1954), 20.4±0.53°C SE recorded in 2004-2007 (Perissinotto et al., 2010) are recorded during the winter months of June-August while the highest mean temperatures e.g. >27°C (Day, 1954), 29.1±0.5°C SE recorded in 2004-2007 (Perissinotto et al., 2010) are recorded in the summer months of November-February. However values greater than 50°C have been recorded in the system during adverse drought conditions (Muir & Perissinotto, 2011).

The northern embayment generally has a wider range of temperature due to its shallower nature as the water rapidly warms up and cools down(Johnson, 1977) e.g. 17.4-36.6°C in 2004- 2005 (Perissinotto et al., 2010). The relatively deeper Channel and the Mouth have a lesser range of temperature variations e.g.

18.6-32.3°C (Perissinotto et al., 2010). This is not only because deeper waters do not easily give up their energy (heat), but also due to tidal influence during the open mouth phase on these parts as the sea experiences much smaller temperature fluctuations (Johnson, 1977). Little variations have been recorded between the temperature at the surface and the bottom of the system suggesting that the system is well mixed.

1.3.3.5 Dissolved Oxygen

Dissolved oxygen in the St. Lucia estuarine lake system is highly variable throughout the year with no defined seasonal trend (Pillay & Perissinotto 2009). Along the estuarine system, dissolved oxygen correlates positively with depth, however, it reduces with increase distance from the mouth. Hence highest (e.g. 5.4-12.8 mg L^{-1}) values are recorded in the Channel and least values (e.g. 0.1-12.6 mg L^{-1}) in the northern embayments (Pillay & Perissinotto 2009).

1.3.3.6 Nutrients

Overall, there is not much knowledge about the nutrient conditions in Lake St Lucia. During the closed mouth phase of the St. Lucia estuarine system in 2007, dissolved inorganic phosphorus (DIP) was limiting whilst dissolved inorganic nitrogen (DIN) was not, hence the high DIN:DIP ratio (van der Molen & Perissinotto, 2011). DIN:DIP ratios decreased after mouth opening events suggesting that turbulence from the tidal flow re-suspends DIP from the bottom sediments at the mouth and Channel (van der Molen & Perissinotto, 2011). In 1975, phosphates had been recorded to increase from July (10µg/L) to December (41µg/L) in False Bay while ammonia and nitrates were high in autumn (170µg/L), reduced in winter (70µg/L) and increased again in October. This suggests nutrients were not limiting in False Bay (Day, 1981).

1.3.4 Phytoplankton

Only few studies have investigated the 'pelagic' phytoplankton of the St. Lucia estuarine system. Cholnoky (1968) surveyed the taxonomy of diatoms, Johnson (1977) conducted a study on phytoplankton and Fielding et al. (1991) studied algal biomass (chl a). These three studies were conducted when the inlet was open. Recently, Bate & Smailes (2008) surveyed diatoms of the system, Perissinotto et al. (2010)studied microalgal biomass, Muir & Perissinotto(2011) investigated the cyanobacteria bloom in 2009/2010, phytoplankton productivity was investigated by van der Molen & Perissinotto (2011) and microalgae (chl a) dynamics by Tirok & Scharler (2013). These recent studies were carried out during the drought periods when the estuarine mouth was closed with the exception of Perissinotto et al. (2010) whose study was carried out when the mouth was closed and briefly opened in 2007.

Diatoms are the dominating group of phytoplankton in the St. Lucia estuarine system. However, nanoplanktonic dinoflagellates, Cyanophyceae, flagellates, and Chlorophyceae have also been recorded (Johnson, 1977). The abundance of phytoplankton is directly proportional to salinity (Johnson, 1977; Bate & Smailes, 2008; Pillay & Perissinotto, 2009). This is evident as the abundance of phytoplankton reduced during the onset of rains which reduced the salinity of the system (Johnson, 1977) and under hypersaline conditions of >220, a *Cyanothece* bloom occurred in 2009-2010 (Muir & Perissinotto 2011). The St. Lucia estuarine system is said to be dominated by euryhaline marine species though a few brackish and ultrahaline species were recorded (Johnson, 1977). Johnson (1977) sampled just after the 1968-1972 droughts and the system was artificially connected to the sea (Taylor, 2006) hence there was an influx of marine species, which is the reason why the euryhaline marine species dominated. Unfortunately, no other studies have looked at phytoplankton as a whole in the system since 1977 and this study is the first to do so. However from diatom studies by Bate & Smailes (2008), 50% of the diatoms encountered were from freshwater sources. This suggests that although some phytoplankton are brackish (estuarine) in nature, riverine and marine water influx to the system also influences the community composition in the system. With the exception of a few centric diatom species such as *Cyclotella* sp. recorded by Cholnoky (1968),Bate & Smailes (2008) did not record any other pure planktonic species in the system during their studies suggesting that the diatom taxa were benthic. In the system, the phytoplankton diversity differed at the same site with time (Bate & Smailes, 2008).

1.4 Aim

Generally, changes in phytoplankton species composition strongly influence the biomass and community structure of higher trophic levels as well as various ecosystem processes however, little is known about the phytoplankton community structure and dynamics in the St. Lucia estuarine system during the extended drought periods when the estuarine mouth was closed. This study thus seeks to characterize the nano- and microplankton (autotrophic and heterotrophic) at different sites of the lakes and investigate their temporal changes under the influence of changing physico-chemical conditions. In St. Lucia, the probability of occurrence of extreme environmental conditions (droughts and floods) is high (Forbes & Cyrus, 1993). Therefore an increase in the knowledge of the effects of these varying physico-chemical conditions on the nano- and microplankton of the system under these extreme conditions of drought experienced during the study will give information on how nano- microplankton is affected. This information will be used to help predict changes in environmental water quality and predict fisheries potential from biomass and species composition in the estuarine ecosystem. Nano- and microplankton are thus of major importance in these ecosystems.

1.5 Objectives

The objectives of this study were

1) To understand and establish a trend in the changes of the nano- and microplankton community and abundance with the varying physico-chemical characteristics.
2) To estimate the biomass (carbon) of nano- and microplankton from cell counts and biovolume measurements.
3) To compare with and add onto Johnson's (1977) list of phytoplankton of Lake St. Lucia.

The main questions addressed in this study were

1) Is there a difference in the nano- and microplankton community between the different compartments of the lake and between seasons?
2) What is the relationship between the nano- and microplankton community structure (composition, abundance, biovolume and biomass) and physico- chemical variables?

CHAPTER 2

MATERIALS AND METHODS

2.1 Sampling

The area of the St. Lucia estuarine lake system is 300-350km^2 (Begg, 1978). Considering its vast surface area, sample sites had to be chosen to best represent the system. Bearing in mind that this system is subdivided into 3 main compartments: northern embayments, South Lake and the Channel which terminates in the mouth, a site was chosen from each compartment. This was possible because Johnson (1977) examined 4 sites in False Bay, 6 sites in North Lake, 4 in the South Lake and 4 in the Channel in 1973 and came to the conclusion that there is a degree of homogeneity within a compartment. Furthermore, boats could not be used due to the very shallow nature of the lake in 2010/2011. Hence emphasis was laid on sites that could be accessed throughout the study period. Three sites were thus chosen for this project. Lister's Point (LP) was sampled for False Bay, which was considered to be representative of the northern embayments. Charters Creek (CC) was sampled as a representative for South Lake and the Mouth (MT) was sampled to represent the lower part of the Channel. For this study the remainder of the Channel was not included due to time.

Since the water depth at Lister's Point, Charters Creek and most times at the Mouth was less than one meter deep, only subsurface samples (<0.5 m depth) were collected and analysed. Also, the St. Lucia estuarine system, has a high degree of vertical mixing (van der Molen & Perissinotto, 2011) and no prolonged vertical stratification (Day, 1954), possibly due to the high turbidity, the wide and shallow nature of the system especially in Charters Creek and Lister's Point. Hence no difference was expected between the surface and bottom samples. Also, studies before now have not shown a difference between the surface and bottom samples of this system (Johnson, 1977). Therefore only sub surface samples were analysed in this study.

Sampling was conducted monthly from October 2010 – September 2011 by K. Tirok (Tirok & Scharler, 2013). Phytoplankton samples were collected in 250ml acid washed iodine proof polyethylene bottles. For nutrients and total autotrophic pelagic biomass (chl a) measurements, samples were collected in 1000 ml polythene bottles. After collection, phytoplankton samples were immediately fixed with acid Lugol's solution, the other samples were kept in the dark to prevent further reactions such as grazing in the collected sample. I took part in subsequent survey trips to St, Lucia where similar samples were collected and have thus familiarised myself with the methodology.

2.2 Physico-chemical measurements

During sampling at each site, salinity, temperature (□C) and turbidity (NTU) were measured using a YSI 6920 Multiprobe system. Depth (m) of LP and CC were measured using a ruler. There were no representative depth measurements for the Mouth because sampling was conducted from the shore, but total depth usually exceeded 2 m. Average depth of MT was estimated at 2 m (Tirok and Scharler 2013). Photosynthetic available radiation (PAR) (µmol photons m^{-2} s^{-1}) was measured at the surface and bottom of all sites using a LI-COR light meter, fitted either with LI-189 or LI- 193SA quantum sensor. The light attenuation coefficient was calculated using the following formula:

$$Kd = \frac{-Ln\left(\frac{Iz2}{Iz1}\right)}{(z2-z1)}.$$

Here, Iz2 is the irradiance (µmol photons m^{-2} s^{-1}) at depth (m) z2 (surface) and Iz1 was the irradiance at depth (m) z1 (bottom).

For dissolved inorganic nutrients, surface water samples were collected in 100 ml bottles, filtered through a Whatman GF/F glass- fiber filter and stored at <-20 □C until analysed. A Skalar San ++ Automated Wet Chemistry Analyser with a 1050 sampler was used to measure dissolved inorganic nitrogen (DIN: nitrate, nitrite and ammonia) and dissolved inorganic phosphorus (DIP: orthophosphate). For chl a determination, known volumes of water samples were filtered through 20 µm Nitex and 2.0 µm Millipore filters for microplankton and nanoplankton size fractions of chl a respectively. Chlorophyll a was measured using the non-acidification method, following (Welschmeyer, 1994). Samples were extracted over 12 to 24 hours in 90% acetone at <5°C in the dark. A Turner Designs fluorometer (10-AU) fitted with narrow band filters was used to measure chlorophyll-a (436 nm) concentrations.

2.3 Nano- and microplankton community composition and biomass

2.3.1 Preservation of nano-and microplankton samples

Samples collected in the 250ml bottles were fixed immediately after collection with 1ml of 2% acidic Lugol's solution [2g of KI, 1g of Iodine, 20ml glacial acetic acid in 200ml of distilled water(National Research Council, 1969)]. This fixative adds weight to the nano- and microplankton cells by discharging any gas that might be held in the vacuoles of the cells, which is and additional advantage during settling (Lund et al., 1958). The fixative stops all biological processes such as grazing in the sample and preserves the cells avoiding further breakage or disintegration. Acidic Lugol's solution also acts as a stain, easing the view of cells under the microscope. It should be noted that, although sampling was done in 2010/2011 and analysis were only done in March 2012–March 2013. Acidic Lugol's fixative is said to preserve

samples for a year however, samples were not disintegrated after two years as samples collected in October 2010 (first sampling date) showed little or no disintegration when analysed in October 2012. Problems with long term storage of plankton samples include the loss of flagella by some flagellated species making identification difficult and contamination with bacteria over time (National Research Council, 1969). Altogether, all samples analysed did not show major problems thus not hindering the identification (to genus level), counts for abundance and biovolume measurements of the cells.

2.3.2 Utermöhl method

In order to calculate the abundance of nano- and microplankton in estuarine waters, a known volume (2-150 ml on the density of cells and detritus in the sample) of sample was analysed for cell identification and counts. Many methods could be used to determine the community composition of nano- and microplankton cells. The use of settling chambers is a good way of concentrating cells of a known volume. However, one of the challenges involving community composition and structure determination of nano- and microplankton is using a subsample that best represents the site in study. The Utermöhl method is preferable as the samples are mixed properly and subsamples poured into the settling chamber directly from sample bottles. This gives less room for errors based on subsample selection. Also, nano-and microplankton identification and photographing of cells are much easier in the settled samples. However, the settling method is not preferred for live samples as live flagellates and cyanobacteria do not readily settle down. The Utermöhl settling method which is probably the best known method for quantitative phytoplankton enumeration (National Research Council, 1969; Paxinos & Mitchell, 2000) was then used for this study bearing in mind that preserved samples were used.

The Utermöhl chamber was used. The chamber is accompanied by settling cylinders of different volumes, namely 2-100ml. The volume to be settled determined the volume of the cylinder used while the density of the cells in the sample determined the volume to be settled. Smaller volumes were settled for more dense samples hence smaller cylinders were used. It was assumed that samples with high chl a values had more cells which proved to be true. The volume of sample settled not only depended on the abundance of cells in the sample but also on the amount of detritus and sediment in the sample (National Research Council, 1969). Samples with more detritus appeared darker than samples with less detritus. For example, small aliquots from samples of CC were settled because CC had a high amount of sediment and/or detritus. This way excessive density of cells and or detritus on the cover slip (chamber floor) was prevented. This was to ensure proper identification and enumeration of cells of all sizes as larger cells are quite visible even in a crowded chamber but small cells of less than 5 µm are easily missed or not seen at all in crowded chambers. This was of interest as estuaries contain high amounts of particulate matter

making microscopy analysis very laborious and limited in its resolution (Azémar et al., 2007) and St. Lucia estuarine lake has high turbidity.

The principle the settling chambers uses is that nano- and microplankton cells will be denser than water hence settle at the bottom of the chamber after a specific period of time (National Research Council, 1969; Paxinos &Mitchell, 2000). Convection currents within the chambers may prevent some cells from settling (Hasle, 1978). This has been shown to be minimal or absent in the towers of volume 2-50ml which were mostly used in this study. However, in the 100ml towers, the convection current may be very applicable but minimised if the sample is allowed to settle for 48hours and the whole chamber counted (Hasle, 1978).The settling time is overall dependent on volume settled. Two milliliters was settled at least for 3 hours, 10 ml for 8 hrs, and 50ml for 24 hrs. Using these values, the time for settling intermediate volumes was extrapolated by a simple linear equation;

$$Time\ (hrs) = \frac{50}{24} * X\ (ml)$$

Here, X is the volume of sample to be settled in milliliters.

2.3.3 Identification and abundance of nano- and microplankton

In aquatic ecology, an understanding of the diversity of microscopic life is of utmost importance when assessing habitat carrying capacity, water quality and trophic web stability. Therefore, a direct microscopic observation, enumeration and documentation of species composition and biomass are the only means of assessing morphological changes within an algae community as a respond to physico-chemical variables.

A volume of 2-100 ml was settled and viewed under a Nikon *ECLIPSE Ti Series* inverted microscope fitted with a DS-US camera powered by the NIS-Elements BR software at magnifications of 100x-400x. Cells were identified to the lowest taxonomic level possible with the aid of taxonomic guides which included Round et al. (1990) for diatoms, Rippka et al. (1979) for cyanobacteria, Tomas (1997) for the other phytoplankton and Post et al. (1983) and Lynn (2010) for ciliates. Transects, fields of view or grids (using the NIS-Elements BR software) were counted for abundant taxa and the whole chamber was counted for rare taxa and large cells (>50 µm). The whole chamber was again always scanned to ensure all taxa were recorded. According to Hillebrand et al. (1999), in order to reduce statistical error during the abundance calculations, at least 50 cells per taxon should be counted and a sum of at least 500cells should be counted per sample. Thus, subsamples of each sample was settled and counted until at least 50cells per

taxa or 500cells per sample were counted and until no new taxa were found. Abundance (Cells per L) was calculated as:

$$Abundance\ (Cells/L) = \frac{A\ (mm2)*1000}{a\ (mm2)*V\ (ml)} * n$$

Here, n is the number of cells counted, A is the area of the whole chamber, a is the area counted, V is the volume settled. The main groups identified and measured included diatoms, cyanobacteria, dinoflagellates, green algae and ciliates. For taxa that were not found in a sample, a zero replacement value was used. A zero replacement value was used on the assumption that if the whole system was analysed (which is impossible) the taxon might be found. The taxon might be present but at densities that cannot be detected in the samples collected. The replacement value was half the detection limit of the taxa. The detection limit was gotten by assuming that 1cell of the taxa was found in the sample.$Detection\ limit\ per\ liter = \frac{1}{X} * 1000$.

Here, X= volume settled in millilitres.

$$Zero\ replacement\ per\ liter = Detection\ limit\ per\ liter * \frac{1}{2}$$

2.3.4 Biovolume

For cell volume calculations, the linear dimensions (length, width, at times height) of at least 25 cells per taxa per sample were measured using the NIS-Elements BR software. These linear dimensions were fitted into geometric models (Table 1) according to Hillebrand et al. (1999), Sun (2003) and Lecce-Monteroni (2007) in order to calculate the cell volume. The shapes for all main groups except for diatoms needed two linear dimensions (length and width) easily measured with the microscope. For diatoms, in addition to the two linear measurements a 3rd dimension (height) was needed for the geometric models. The girdle view (length and height) of most diatom species were hardly seen hence the height could not be measured. A relationship (linear regression) between the heights and widths of some species in my samples and already recorded dimensions (width and height) in the Helsinki Commission of 2006 was established. The regression equation of species such as *Gyrosigma* sp. and *Suirella* sp. whose r^2 value was 0.832 and 0.930 respectively was then used in this study. Measured widths were thus computed into the regression equations ($y = 0.778x + 0.669$ for *Suirella* and $y = 0.643x + 1.25$ for *Gyrosigma* where, y was cell height and x was the width of the cell). Also, the following assumptions were made: for taxa whose longest dimension was <20µm, the height was equal to the width (Helsinki Commission, 2006), for

Nitzschia species with a width of <20µm, the height was equal to the width(Helsinki Commission. 2006)and for other species with a width >20µm, height was equal to half the width (http://oceandatacenter.ucsc.edu/PhytoGallery/phytolist.html).Using these assumptions, the cell biovolume for diatoms was calculated. Total biovolume per taxon per liter was calculated by multiplying the mean cell biovolume for each taxon by the abundance of that taxon per liter (Felip & Catalan, 2000). Zero replacement values for biovolume per liter were calculated by multiplying the abundance zero replacement value of the taxon by the least cell volume of that taxon in another sample.

Table 1: Abundant taxa, shape and geometric equations for cell volume. Here, d = diameter, h = height and l = length and z = length of cone on the sphere (z only applies for *Didinium* and *Fabrea* ciliate taxa).

Main Group	**Taxa**	**Shape**	**Geometric equation**
Green algae	*Chlamydomonas*	Prolate spheriod	$\pi/6 \cdot d^2 \cdot h$
	Dunaliella	Prolate spheriod	$\pi/6 \cdot d^2 \cdot h$
	Chroomonas	Prolate spheriod	$\pi/6 \cdot d^2 \cdot h$
Dinoflagellates	All encountered except *Protoperidium*	Prolate spheriod	$\pi/6 \cdot d^2 \cdot h$
	Protoperidium	Cone + ½ sphere	$\pi/12 \cdot h^2 \cdot (d+z)$
Ciliates	All encountered except *Didinium, Fabrea* and *Laboea* .	Prolate spheriod	$\pi/6 \cdot d^2 \cdot h$
	Didinium and *Fabrea*	Cone + ½ sphere	$\pi/12 \cdot h^2 \cdot (d+z)$
	Laboea	Cone	$\pi/12 \cdot h^2 \cdot l$
Cyanobacteria	*Cyanothece*	Sphere	$\pi/6 \cdot d^3$
	Cyanothece	Prolate spheriod	$\pi/6 \cdot d^2 \cdot h$
	All filamentous forms encountered	Cylinder	$\pi/4 \cdot d^2 \cdot h$
Diatoms	*Navicula* , *Nitzschia* , *Diploneis* , *Gyrosigma* , *Entomoneis* , *Triblionella*	Eliptic prism/ prism on parallelogram base	$½ \cdot l \cdot d \cdot h$
	Triceratiaceae, Heliopeltaceae, Melosiraceae	Cylinder	$\pi/4 \cdot d^2 \cdot h$
	Amphora , *Seminavis*	Cymbelliod	$(\pi/6 \cdot l \cdot d \cdot h) - 35\%$

2.3.5 Biomass

The biovolume of phytoplankton groups was converted to biomass [pg C per liter] by the use of allometric relationships according to Montagnes et al. (1994) and Gosselain (2000). These conversion factors took into consideration the different taxa. The equation used was $y = a \cdot x^b$ where y is the carbon (pg/µm^3), x is the biovolume (µm^3), a (y-intercept of regression equation) and b (slope of regression equation) are constants (Gosselain, 2000). Different set of constants (a, b) were used for diatoms and another set for groups other than diatoms. However, the same constants were used for all cell sizes in each case (diatoms and non- diatoms) because all individual cell biovolume were within the range 10^0-10^6 µm^3 (Table 2) (Menden-Deuer & Lessard, 2000).

For ciliates, the amount of carbon was determined by using the C:biovolume ratio of 0.19 pg/μm^3 which applied to ciliates of all cell sizes (Putt & Stoecker, 1989). It has been noted that fixation by acid Lugol's solution causes nano- and microplankton cells to shrink and thus has an effect on the biovolume and hence on the estimated carbon content. The effect of shrinking up of cells due to the acid Lugol's solution was rectified by using constants (Table 2) determined from cells fixed in acid Lugol's solution where the effects of the distortion in cell size was already considered.

Table 2: Constants used in the calculation of carbon for diatoms and non-diatoms cell volume (biovolume of a single cell) preserved in Lugol's solution.

Reference	*a*	*b*	Main group	Sample size	Cell volume (μm^3)
Menden-Deuer & Lessard, 2000	0.288	0.811	Diatoms	94	10^0- 10^6
	0.216	0.939	Non- diatoms	91	10^0- 10^6

Chlorophyll a values (mg/ m^3) for the nano- and microplankton size class, were converted to biomass (carbon) pg/L by assuming that the ratio of chl a:C is 1:50(Reynolds 2006). A linear regression was then conducted to establish a relationship between the biomasses from both methods. Also, a chl a (from the nano- and microplankton size class):C (carbon from phytoplankton only) ratio was determined.

2.4 Data analysis

In order to determine the association between the biological variables [dependent variables: chl a, abundance, biovolume, biomass (carbon)] and independent variables which were sampling seasons and physico-chemical variables (turbidity, salinity, depth, temperature, light attenuation coefficient (Kd) and nutrients), a multiple linear regression analysis was performed using IBM SPSS 21. The output of this analysis also gave Pearson correlation coefficient between the biological variable and the independent variables. Abundance, biovolume and biomass data were log transformed before the analysis to ensure normal distribution of data. A one-sample Kolmogorov-Smirnov test was then conducted on the residuals to confirm normality in the distribution of the data. In all statistical analysis performed, the degrees of freedom (*df*) was 37 and the total sample size (n) was 38.

In order to obtain a visual perspective of the relationship between samples in terms of abundance, biovolume and biomass per site and season, non-metric multidimensional scaling (nMDS) plots were

produced using PRIMER (Plymouth Routines In Multivariate Ecological Research) 6 package. Data were fourth root transformed and a Bray-Curtis similarity measure was produced to yield a resemblance matrix. An nMDS plot was then drawn using the resemblance matrix such that biomass of communities clustered based on their similarity and the distance between taxa is determined by how dissimilar the taxa were (Anderson et al., 2008). The nMDS plots were drawn for all nano-and microplankton taxa with respect to site and seasons.

R (R core team 2013) with package vegan (Oksanen et al., 2013) was used to visualise the relationship between the physico-chemical variables and the abundance of the dominant abundant taxa using Canonical Correspondence Analysis (CCA). The ordination axes used were weight sums of the physico-chemical variables formed by CCA constrained ordinations. Two dimensional plots were used to represent the results. The longer the distance of the physico-chemical variables from the center of the plot, the more important the variable was and the position of the taxa relative to the variable indicates the correlation such that the closer the taxon is to the variable, the stronger the relationship is. Statistics for the first five axes were presented. Taxa abundance were square root transformed before analysed.

In order to determine if there were differences in the nano- and microplankton groups between sites and seasons, a Permutational Analysis of Variance (PERMANOVA) using PRIMER 6, was then conducted. The hypothesis tested was that there was no difference between sites and seasons. All data were fourth root transformed and a S17 Bray-Curtis similarity measure was done to analyse for differences in community structure between site and seasons. In creating the PERMANOVA design, no nested factors were chosen, factors to be analysed were considered fixed and no specific contrast was specified. The permutation method used was an unrestricted permutation of raw data and 999 permutations were conducted.

CHAPTER 3

RESULTS

3.1 Physico-chemical characteristics and chl a

There was no connection with the sea during this study as the mouth was closed. Salinity in the system during this study ranged from 4.62- 157.5. Salinity in LP in the northern embayments varied highly from 43.20- 157.32 while the southern Lake ranged from 8.55- 63.65 and MT from 4.62- 22.44. Salinities decreased with the onset of rains in the summer months of 2011 till the end of study in the system. In LP, salinities decreased from 92.60 in the spring month of October 2010 to 46.35 with the onset of the rains in January 2011 (Figure 2).Salinity differed significantly (p<0.001, F= 12.75, n= 37) between sites. The system, most especially the northern embayment was hypersaline and had a reverse salinity gradient. The onset of rains also led to a slight increase in the depth of the shallow system which ranged from 0.06-0.45 m in LP, approximately 2m and 0.02-0.22 m in MT and CC respectively. Depth significantly (p<0.001, F= 2008.93) differed between sites. Temperatures were generally high ranging from 15.57-34.76 in LP, 16.48- 29.45 in MT and 15.43- 30.17 in CC (Figure 2). Lowest temperature values were recorded in winter and highest in summer however, temperature was not significantly (p= 0.38, F= 0.99) different between sites. Turbidity showed no seasonal trend and was not significantly (p= 0.698, F= 0.36) different between sites. Turbidity was highest and highly variable in CC ranging from 13.00- 465.40 NTU, followed by LP ranging from 15.10- 254.20 NTU and lowest in MT ranging from 13.00- 240.80 NTU (Figure 2).

Throughout the study period, DIP ranged from 0.47- 6.34 µM with the highest values of 1.00- 6.34 µM in LP, 0.47- 4.06 µM and 0.84- 3.90 µM in MT and CC respectively. DIN ranged from 0.19- 30.93 µM with highest values of 1.45- 30.93 µM in MT, 0.62- 13.27 µM in CC and the lowest values of 0.19- 9.65 µM in LP. There was no significant (p= 0.93, F= 0.78, n= 37 and p= 0.09, F= 2.59, n= 37) difference in DIP and DIN respectively between the sites. The total N:P ratio ranged between 0.03- 8.96 which is less than the Redfield value of 16 ratio.

Chlorophyll a values during this study ranged from 0.46- 30.08 mg chl a/m^3. The highest values 3.52-30.08 mg chl a/m^3 with a mean of 19.66 mg chl a/m^3 were recorded in LP, 2.59- 11.84 mg chl a/$m^3$with a mean of 4.17 mg chl a/m^3 in MT and the lowest values 0.46- 4.93 mg chl a/m^3 with a mean of 2.85 mg chl a/m^3 in CC. No clear seasonal trend was observed in chl a values (p= 0.474, F= 0.855) however, there were significant (p<0.001, F= 13.16) differences in chl a between sites.

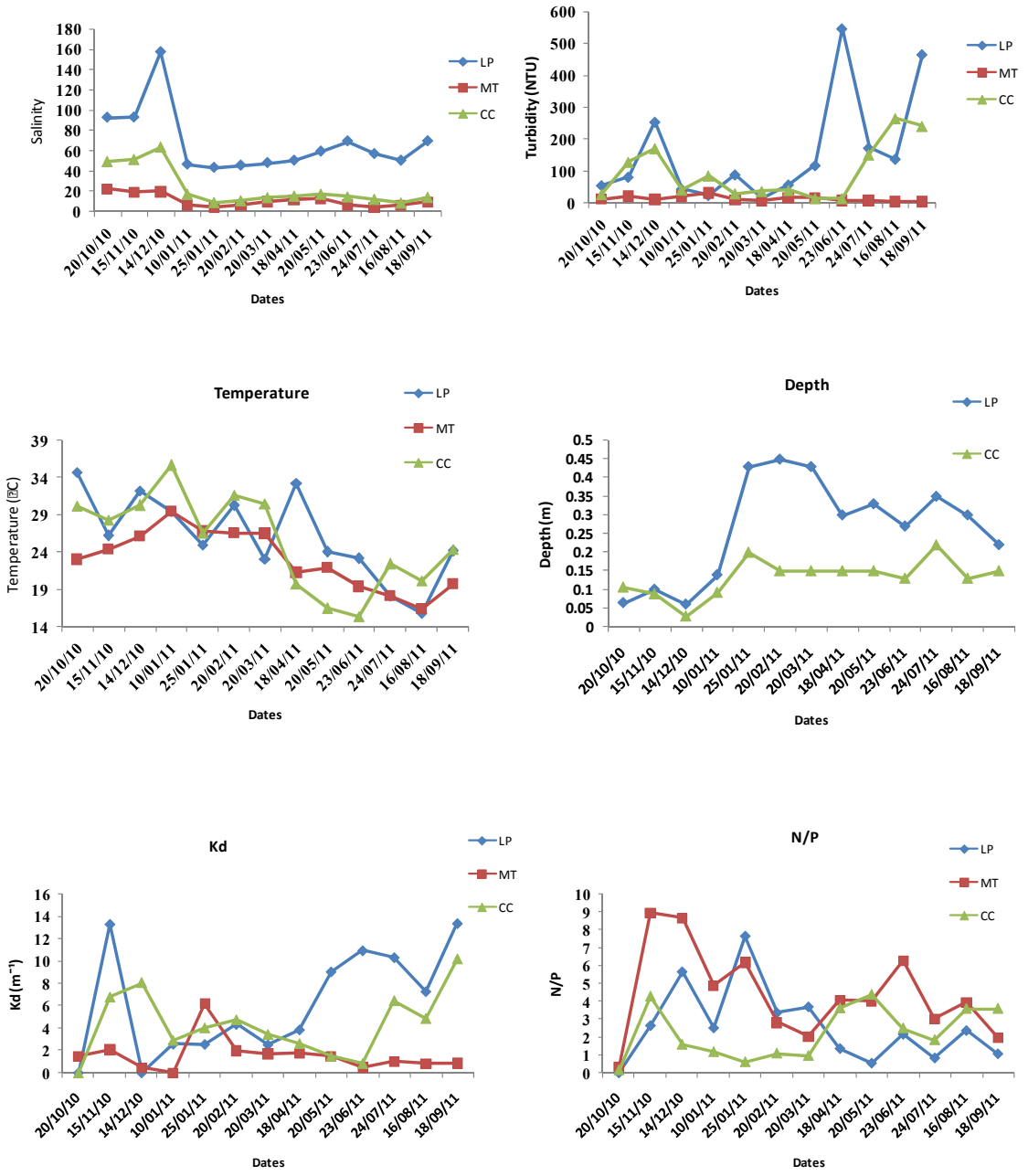

Figure 2: Physico-chemical variables measured during the sampling period at the three stations (LP, MT and CC). Salinity, turbidity, temperature, Kd: light attenuation coefficient, N/P: ratio of nitrogen and phosphorus were available from all three stations. Representative depth measurements were only available for LP and CC but not for MT (see methods for details).

3.2 Nano-and microplankton dynamics

3.2.1 Phytoplankton community composition

Seventy eight phytoplankton taxa were recorded during this study, fifty six of which were diatoms, eight green algae, one cryptophyte, seven cyanobacteria and six dinoflagellates (Table 3 and 4). Two of the

green algae taxa were unidentified flagellates. The eight green algae genera belonged to six families and five orders. *Chlamydomonas* was the main taxon and occurred in all sites and seasons. The two unknown taxa and *Micromonas* only occurred in LP in spring and summer. The genus *Dunaliella* known to flourish and adapt well in high salinities was recorded only in LP during the spring and summer months. The fresh water genera *Kirchneria* and *Scenedesmus* were recorded only in MT and CC. Cryptophytes taxonomically distinct from chlorophytes, similar in morphological and ecological characteristics (Tomas, 1997, Reynolds, 2006) were represented by a single taxon (*Chroomonas*). *Chroomonas* was recorded at all seasons in CC and MT but only occurred in LP during spring.

Of the dinoflagellate taxa recorded, four were armoured and two were naked dinoflagellate genera. One of the taxa was armoured and unidentified. This unknown taxon was only recorded in MT during spring (September 2010). *Protoperidinium*, a heterotrophic armoured dinoflagellate was recorded during all season at LP and MT except during spring at LP. However, this genus also occurred in a very low abundance (75 Cells/L) in CC during summer. The unarmoured heterotrophic species *Oxyrrhis marina* was recorded in LP and MT during all seasons and also in a very low density (37 Cells/L) in CC during spring. The naked *Gymnodinium* was recorded during spring at LP and during autumn at MT and CC. The armoured autotroph *Prorocentrum* occurred only in MT during summer, autumn and winter.

Seven cyanobacteria taxa were identified belonging to five orders. A unicellular genus, one colonial and five filamentous cyanobacteria taxa were recorded during this study. The unicellular *cyanothece* occurred in LP during all seasons while the filamentous taxa *Oscillatoria*, *Spirulina*, and *Lyngbya* occurred throughout the study. Anabaena a filamentous cyanobacterium was recorded only at MT in spring while Nostocaceae recorded at all sites but not during all seasons. The colonial cyanobacterium, *Merismopodia* was recorded only in MT in summer and autumn.

In 2010-2011, 27 diatom genera were identified in the St. Lucia estuarine lake system. These genera belonged to 19 families and 14 orders. Three taxa were unidentified. The genera with a high number of species were *Navicula* with five species and *Nitzschia* with ten species. In general, diatoms occurred throughout this study at all sites during all seasons however, there was a difference in the species composition and abundance between sites and dates. In LP, only the genera *Pleurosigma*, *Gyrosigma*, and *Entomoneis* occurred during all seasons. At MT during spring, only *Diploneis, Craticula*, and the centric diatom *Cyclotella* were recorded. However, centric diatoms *Cyclotella* and *Actinoptychus*, *Amphora*, *Seminavis*, *Nitzschia longissima* and other *Nitzschia* species occurred in all the other seasons at MT. In CC, the diatoms *Amphora*, *Seminavis*, *N. longissima*, *Gyrosigma*, *Diploneis*, *Entomoneis* and the centric

diatoms *Auliscus*, *Actinoptychus*, *Melosira* and *Cerataulina* occurred in during all seasons. At LP, only one centric genus, *Chaetoceros*, was recorded at a very low abundance of 500 Cells/L and it only occurred in autumn. In MT only two centric genera; were recorded and in CC, six centric genera were recorded. The number of centric genera was thus higher in the South Lake than in the Mouth and in the False Bay. Only 12 of the 27 diatom taxa identified during this study were identified in the St. Lucia system in 1973-1975 by Johnson (1977) (Table 3). None of the genera recorded for the other phytoplankton groups during this study were recorded by Johnson (1977).

Table 3: List of diatom genera, families and orders identified from the St. Lucia system in 2010-2011and the sites they were found at. The asterisk (*) represents genera found in the system in 1973-1975 (Johnson, 1977), (^{b}) = benthic taxon, (^{p}) = pelagic taxa, (^{bp}) = a benthic and pelagic taxa, (^{pe}) = pelagic and epiphytic and (^{u}) = unknown.

Genus	Family	Order	Site
Diatoms			
Actinoptychus Ehrenberg 1843*^{b}	Heliopeltaceae	Coscinodiscales	MT,CC
Amphora Ehrenberg, 1902*^{b}	Catenulaceae	Thalassiophysales	LP,MT,CC
Auliscus Ehrenberg 1843*^{b}	Triceratiaceae	Triceratiales	CC
Campylodiscus Ehrenberg, 1844^{b}	Surirellaceae	Surirellales	CC
Cerataulina Schütt, 1896^{p}	Hemiaulaceae	Hemiauliales	CC
Chaetoceros Ehrenberg, 1844*^{p}	Chaetocerotaceae	Chaetocerotales	LP
Cocconies Ehrenberg, 1837^{b}	Cocconeidaceae	Achanthales	MT
Craticula Grunow, 1867^{b}	Stauroneidaceae	Naviculales	MT
Cyclotella Brébisson, 1838*^{p}	Stephanodiscaceae	Thalassiosirales	MT,CC
Diploneis Cleve, 1894*^{b}	Diploneidaceae	Naviculales	LP,MT,CC
Encyonema Kützing, 1833^{b}	Cymbellaceae	Cymbellales	MT
Entomoneis Ehrenberg, 1845^{b}	Entomoneidaceae	Surirellales	LP,MT,CC
Fragilaria Lyngbye, 1819*^{bp}	Fragilariaceae	Fragilariales	MT,CC
Gomphonema Ehrenberg, 1832^{bp}	Gomphonemataceae	Cymbellales	LP,CC
Gyrosigma Hassal, 1845^{b}	Pleurosigmataceae	Naviculales	LP,MT,CC
Hantzschia Grunow, 1877^{b}	Bacillariaceae	Bacillariales	CC
Melosira Agardh, 1824*^{b}	Melosiraceae	Melosirales	MT,CC
Navicula Bory de Saint-Vincent, 1822*^{b}	Naviculaceae	Naviculales	LP,MT,CC
Nitzschia Hassall, 1845*^{bp}	Bacillariaceae	Bacillariales	LP,MT,CC
Odontella Agardh, 1832^{pe}	Triceratiaceae	Triceratiales	CC
Placoneis Mereschkowsky, 1903^{b}	Cymbellaceae	Cymbellales	MT
Plagiodiscus Grunow & Eulenstein, 1867^{u}	Surirellaceae	Surirellales	LP,CC
Pleurosigma W.Smith, 1852*^{bp}	Pleurosigmataceae	Naviculales	LP,MT,CC
Rhopalodia Muller, 1895^{b}	Rhopaliodiaceae	Rhopaliodiales	MT
Seminavis Mann, 1990^{b}	Naviculaceae	Naviculales	LP,MT,CC
Surirella Turpin, 1828*^{b}	Surirellaceae	Surirellales	LP,CC
Tryblionella W.Smith, 1853^{bp}	Bacillariaceae	Bacillariales	MT,CC

Table 4: List of green algae, cryptophyte dinoflagellate and cyanobacteria genera, families and orders identified from the St. Lucia system in 2010-2011and the sites they were found in.

Genus	Family	Order	Site
Green algae			
Chlamydomonas Ehrenberg, 1834	Chlamydomonadaceae	Volvocales	LP,MT,CC
Dunaliella Teodoresco, 1905	Dunaliellaceae	Volvocales	LP
Euglena Ehrenberg, 1838	Euglenaceae	Euglenales	MT,CC
Kirchneria Hindák, 1988	Selenastraceae	Sphaeropleales	MT,CC
Micromonas Manton & Hill, 1960	Halosphaeraceae	Chlorodendrales	LP,MT,CC
Scenedesmus Meyen, 1829	Scenedesmaceae	Sphaeropleales	MT
Unknown flagellate 1			LP
Unknown flagellate 2			LP
Cryptophyte			
Chroomonas Hansgirg, 1885	Cryptomonadaceae	Cryptomonadales	LP,MT,CC
Dinoflagellates			
Alexandrium Halim, 1960	Goniodomataceae	Gonyaulacales	LP,MT,CC
Gymnodinium Stein, 1878	Gymnodiniaceae	Gymnodiniales	LP,MT,CC
Oxyrrhis Dujardin, 1841	Oxyrrhinaceae	Oxyrrhinales	LP,MT,CC
Prorocentrum Ehrenberg, 1833	Prorocentraceae	prorocentrales	MT
Protoperidinium Bergh, 1881	Protoperidiniaceae	Peridiniales	LP,MT,CC
Unknown dinoflagellate			MT
Cyanobacteria			
Anabaena Bornet & Flahault, 1886	Nostocaceae	Nostocales	MT
Cyanothece Komárek, 1976	Cyanobacteriaceae	Chroococcales	LP
Lyngbya Gomont, 1892	Oscillatoriaceae	Oscillatoriales	LP,MT,CC
Merismopodia Meyen, 1839	Merismopediaceae	Synechococcales	MT
Oscillatoria Gomont, 1892	Oscillatoriaceae	Oscillatoriales	LP,MT,CC
Spirulina Gomont, 1892	Spirulinaceae	Chroococcales	LP,MT,CC
Unknown cyanobacteria 1	Nostocaceae	Nostocales	LP,MT,CC

3.2.2 Ciliates community composition

In 2010-2011, 19 ciliate taxa were found. These belonged to nine orders, 11 families and 11 genera (Table 5). Twelve of these taxa were found in LP, nine at MT and eight at CC. The highest number of different species were in the genus was *Euplotes*, a benthic and substrate oriented feeding heterotroph which had six species. This genus occurred mainly at LP during spring. The species *Euplotes* 2 was only recorded at low abundance in winter at MT and at CC during spring. *Fabrea* and *Uronema* occurred in all seasons at LP and were not recorded anywhere else in the system except for MT during autumn when *Fabrea* was recorded with a very low abundance of 190 Cells/L. The tintinnid *Codonella* occurred in MT and CC while *Tintinnopsis* was only recorded in CC. The Choreotrichs occurred throughout this study in all sites

and seasons except for summer in LP and autumn in CC. Genera such as Unknown 1, 3 and 4 were only recorded once at LP during spring, at CC during winter and at MT during summer respectively. (See Plate 1, Appendix, for photographs of selected ciliate taxa).

Table 5: Ciliates occurring in St. Lucia estuarine lake system in 2010-2011.

Genus	Family	Order	Food	Reference	Site
Codonella Haeckel, 1873	Codonellidae	Tintinnida	Bacteria and phytoplankton (<20 µm)	Lynn, 2010	MT, CC
Condylostoma Bory de st. Vincent, 1824	Condylostomatidae	Heterotrichida	Bacteria, microalgae and other protists.	Post et al., 1983	LP,MT,CC
Didinium Stein, 1859	Didiniidae	Haptorida	Flagellates and other ciliates	Lynn, 2010	MT
Euplotes 1 Erenberg, 1831	Euplotidae	Euplotida	Bacteria, microalgae and smaller protistss.	Lynn, 2010	LP
Euplotes 2 Erenberg, 1831	Euplotidae	Euplotida	Bacteria, microalgae and smaller protists.	Lynn, 2010	LP,CC
Euplotes 3 Erenberg, 1831	Euplotidae	Euplotida	Bacteria, microalgae and smaller protists.	Lynn, 2010	LP
Euplotes 4 Erenberg, 1831	Euplotidae	Euplotida	Bacteria, microalgae and smaller protists.	Lynn, 2010	LP,CC
Euplotes 5 Erenberg, 1831	Euplotidae	Euplotida	Bacteria, microalgae and smaller protists.	Lynn, 2010	LP
Fabrea Henneguy, 1890	Climacostomidae	Heterotrichida	Flagellates and smaller protists	Lynn, 2010	LP,MT
Laboea Lohmann, 1908	Strombidiidae	Strombidiida	Bateria, microalgae and smaller protists. Mixotrophic species exist.	Lynn, 2010	MT
Nassula Ehrenberg, 1834	Nassulidae	Nassulida	Bacteria, small protists including microalgae	Lynn, 2010	MT
Strobilidium Schewiakoff, 1893	Strobilidiidae	Choreotrichida	Bacteria and phytoplankton	Lynn, 2010	LP
Strombidinopsis Kent, 1881	Strombidinopsidae	Choreotrichida	Bacteria and phytoplankton	Lynn, 2010	MT,CC
Tintinnopsis Stein, 1867	Codonellidae	Tintinnida	Bacteria and phytoplankton (<20 µm)	Lynn, 2010	LP,MT,CC
Uronema Dujardin, 1841	Uronematidae	Philasterida	Bacteria	Post et al., 1983	LP
Unknown 1	Litonotidae	Pleurostomatida	Flagellates and smaller protists	Lynn, 2010	LP
Unknown 2					CC
Unknown 3	Litonotidae	Pleurostomatida	Flagellates and smaller protists	Lynn, 2010	MT,CC
Unknown 4					LP

3.2.3 Nano- and microplankton abundance

In this study, six main nano- and microplankton groups were found. These groups were diatoms, green algae, cryptophytes dinoflagellates, cyanobacteria and ciliates. The first four groups are referred to as phytoplankton groups. All six groups were found in the estuarine system, however, not all groups were found during each individual sampling session (Figure 3). In this study, taxa were considered abundant if their abundance was ≥ 10% of the total abundance per date and site. The ranges of estimated abundance values of the six main groups have been recorded in table 6.

At LP, cyanobacteria had the highest mean abundance over the entire sampling period of $2.0x10^8$ Cells/L with peaks in the spring months of October – November 2010. This high abundance in cyanobacteria was mostly caused by the *Cyanothece* sp. and to a lesser extent by *Spirulina* sp., *Oscillatoria* sp. and *Lyngbya* sp. Diatoms with a mean abundance of $3.0x10^6$ Cells/L and peaks in the autumn-winter months of April – September 2011 were the next most abundant group in LP. The high abundance in diatoms during the above mentioned season was caused by *Nitzschia* sp.3. In the first three months of the study, green algae, *Dunaliella* sp. and *Chlamydomonas* sp. dominated over diatoms recording a peak abundance of $8.0x10^6$ Cells/L in November 2010. Ciliates had the lowest mean abundance ($1.1x10^5$ Cells/L). Green algae, dinoflagellates and ciliates showed no seasonal trend in their abundance. On the other hand, diatoms increased from October 2010 to July 2011 and remained relatively constant ($1.8x10^6$-$2.1x10^6$ Cells/L) until the end of the study. Cryptophytes were only recorded in September 2011.

At MT, cryptophytes dominated in spring (October and November 2010) after which green algae dominated in the rest of the study, however, there was a significant increase in the mean cyanobacteria abundance in the summer month of February and March. This increase was due to the *Merismopodia* sp. recording an average cell count of $1.34x10^8$ Cells/L. Cyanobacteria was thus the most abundant group in the system recording a mean abundance of $1.20x10^7$ Cells/L while ciliates had the least mean abundance ($5.01x10^3$ Cells/L). The main ciliate taxa at this site were of the subclasses Choreotrichia, order Tintinnida and Choreotrichida. However, at CC unlike at LP and MT, green algae were the most abundant phytoplankton group with a mean abundance of $2.8x10^5$ Cells/L while dinoflagellates were the least ($1.3x10^2$ Cells/L) abundant.

PERMANOVA analysis showed that there was no significant difference ($F=1.1764$, $p= 0.333$) in nano- and microplankton abundance between seasons however, there was a significant difference ($F=5.2041$, $p= 0.002$) in the abundance between sites (Table 7). All five groups were most abundant in LP except for green algae and cryptophytes which were most abundant in MT (Figure 3). The lowest diatom abundance

($1.4x10^5$ Cells/L) was recorded in MT and the lowest cyanobacteria ($4.9x10^3$ Cells/L), green algae ($2.9x10^5$ Cells/L) and dinoflagellate ($1.3x10^2$Cells/L) abundance was recorded in CC (Figure 3). Ciliate abundance was highest in LP ($1.08 x10^5$ Cells/L), followed by MT ($5.01x10^3$ Cells/L) and CC ($1.10x10^3$ Cells/L) (Figure 4).

Table 6: Ranges of abundance (Cells/L) of main nano- and microplankton groups at the three sampling sites. The values with an asterisk (*) are zero replacement values.

Main group/Site	LP	MT	CC
Diatom	$6.00x10^3$-$1.77x10^7$	$1.08x10^2$-$1.38x10^6$	$3.61x10^3$-$1.06x10^6$
Cyanobacteria	$3.36x10^4$-$1.11x10^9$	8.33*-$1.34x10^8$	1.25x10*-$5.30x10^4$
Green algae	2.50x10*-$8.02x10^6$	$4.00x10^2$-$8.02x10^6$	1.25x10*-$1.77x10^6$
Cryptophytes	$2.50x10^2$-$4.60x10^4$	5.00x10*-$6.82x10^5$	1.46x10*-$5.35x10^5$
Dinoflagellate	2.50x10*-$5.84x10^5$	2.50x10*-$3.84x10^5$	8.33*-$9.00x10^2$
Ciliates	$1.63x10^2$-$4.98x10^5$	8.33*-$2.12x10^4$	1.25x10*-$5.53x10^3$

Table 7: F and *p* (perm) values (***= $p<0.001$, strongly significant) from PERMANOVA analysis of abundance, biovolume and biomass of individual taxa between seasons and sites.

	Seasons		Sites
	F	*p*	F
Abundance	1.1764	.333	5.2041***
Biovolume	1.2282	.292	6.1825***
Biomass	6.1825	.29	6.2913***

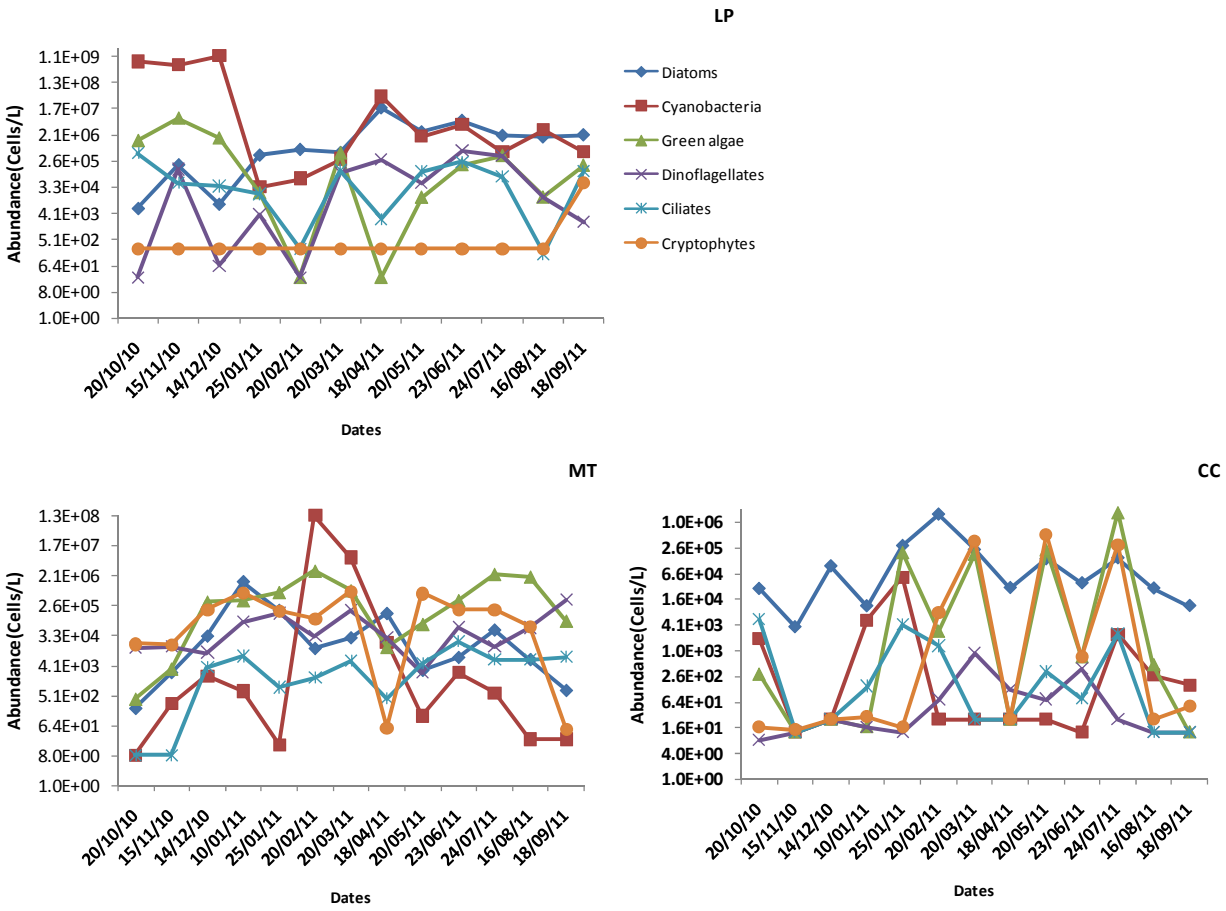

Figure 3: Mean abundance (Cells/L, mean of two replicates per date) of main nano- and microplankton groups in LP, MT and CC.

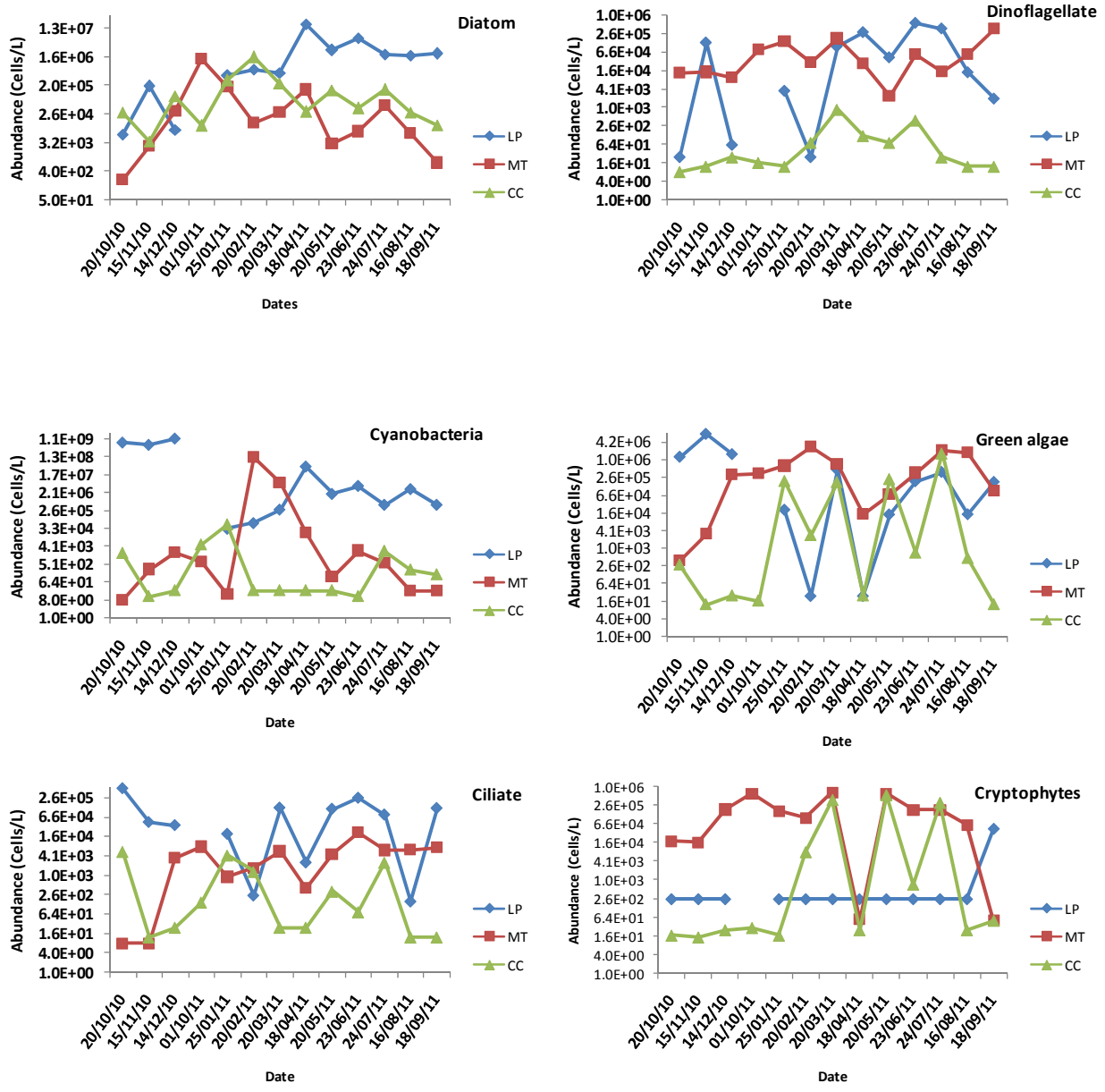

Figure 4: Mean abundance (Cells/L) of all 6 groups in LP, MT and CC. No data were available for LP on the 01/10/11.

3.2.4: Nano- and microplankton diversity and richness

The taxon richness varied with sampling dates and sites. Taxon richness decreased from the estuarine mouth towards the northern embayments. Lister's Point had the least (56) total number of taxa while CC had 63 taxa and MT recorded the highest number of taxa (79). However, the lowest taxon richness (six) per date and site was recorded in CC early in the month of January 2010. With respect to taxon richness per site and date, LP had a median of 16, CC had 18 and MT had 24 (Figure 5). The diversity index however had a different trend to taxon richness. The diversity index was highest in LP with a median of 2

followed by MT with a median of 1.8 and lastly CC (1.3) (Figure 5). It should be noted that LP had a high variability with respect to diversity indices. Although pelagic samples were collected, more than 70% of the diatom taxa identified was benthic in nature. Charters Creek had the highest percentage of pelagic taxa and LP had the lowest (Figure 6).

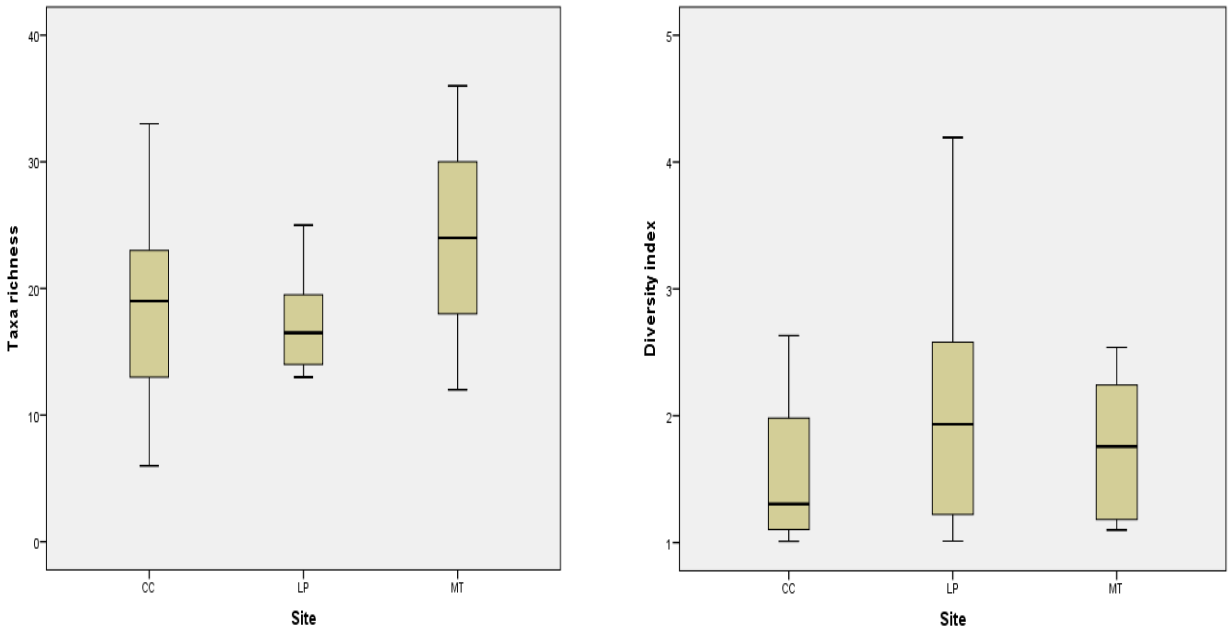

Figure 5: Mean ±2 SD of taxon richness and Shannon-Wiener Diversity of all sampling sessions per each site.

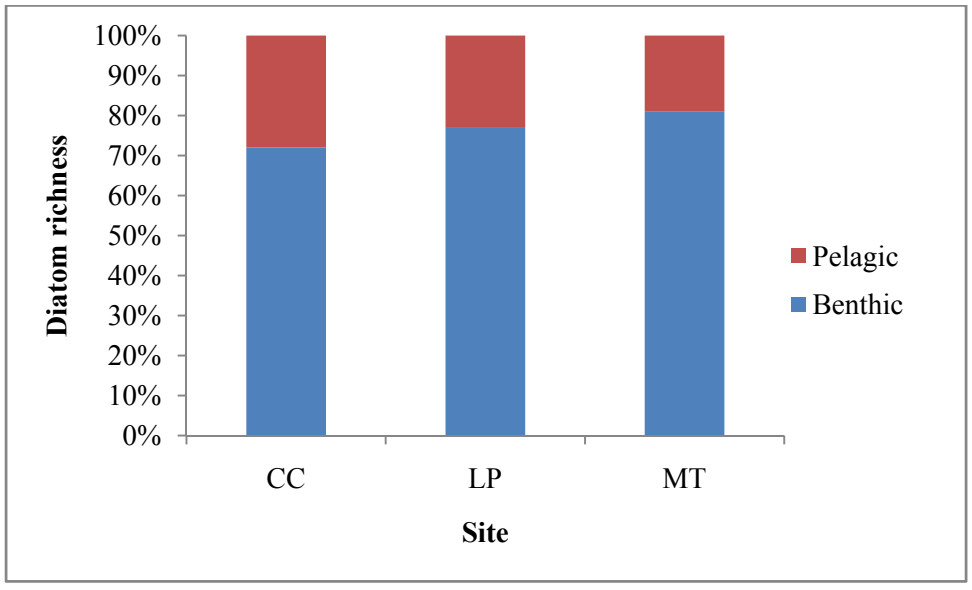

Figure 6: Percentage taxon richness of benthic and pelagic diatom taxa per site.

3.2.5 Dominant nano- and microplankton taxa

Out of the 56 taxa recorded at LP during this study, only 13 of these taxa accounted for more than 10% of the nano- and microplankton abundance and 12 for biomass (Table 8). Of the 79 taxa recorded at MT, only 10 of these taxa accounted for more than 10% of the nano- and microplankton abundance and 13 for biomass (Table 9). All of the diatoms in the dominant abundance taxa in LP and MT were pennate diatoms however, only one diatom *Entomoneis* was present in the biomass dominant taxa in LP and no diatom in MT. Dominant taxa for biomass was made of ciliates and dinoflagellates in LP and green algae, ciliates and dinoflagellates in MT. Only 20 of the 63 taxa recorded at CC during this study, accounted for more than 10% of the nano- and microplankton abundance and 16 for biomass most of which were diatoms. Of the 20 dominant taxa for abundance, four were centric diatoms, four of the genus *Nitzschia* and two of the genus *Gyrosigma*. Hence, these three taxa were the most dominant in CC during this study (Table 10). On average, only four taxa accounted for more than 90% of the nano- and microplankton abundance at LP and CC and two taxa accounted for more than 90% of the abundance at MT (Figure 7).Therefore in the system, although the diversity was high only a few taxa can survive hence dominate at a particular time under the given environmental conditions. Due to the variation of these taxa in size and carbon content, species that dominated in abundance did not necessarily dominate in biomass. Photographs of some of the dominant taxa can be found in Plate 1-3 (Appendix). Table 11 shows the salinity ranges of some of the abundant species recorded in the system by previous studies.

Table 8: Dominant taxa found at LP from October 2010-September 2011 where dominant taxa are defined as those contributing ≥ 10% of the nano- and microplankton abundance and biomass (*).

Month/Site	Listers Point				
Oct-10	*Cyanothece*				
*	*Cyanothece*	*Fabrea salina*	*Spirulina*		
Nov-10	*Cyanothece*				
*	*Cyanothece*	*Spirulina*	*Fabrea salina*		
Dec-10	*Cyanothece*				
*	*Cyanothece*	*Fabrea salina*	*Dunaliella*		
Jan-11	*Nitzschia* 1 and 2	*Nitzschia longissima*			
*	*Uronema*	*Protoperidinium*	*Cyanothece*	Unknown ciliate 4	
Feb-11	*Navicula* 1	*Navicula* 2	*Nitzschia sigma small*		
*	Nostocaceae	*Lyngbya*	Unknown ciliate 4		
Mar-11	*Cyanothece*	*Chlamydomonas*	*N. sigma small*		
*	*Fabrea salina*				
Apr-11	*Cyanothece*	*Nitzschia* 3			
*	*Cyanothece*	*Fabrea salina*			
May-11	*Cyanothece*	*Nitzschia* 3			
*	*Strobilidium*	*Fabrea salina*			
Jun-11	*Cyanothece*	*Navicula* , nbl	*Nitzschia* 3		
*	*Strobilidium*	*Oxyrrhis marina*	*Cyanothece*		
Jul-11	*Cyanothece*	*Chlamydomonas*	*Oxyrrhis marina*	*Nitzschia* 3	*Nitzschia* dcl
*	*Strobilidium*	*Oxyrrhis marina*	*Fabrea salina*		
Aug-11	*Cyanothece*	*Nitzschia* 3	*Nitzschia* dcl		
*	*Oxyrrhis marina*	*Cyanothece*			
Sep-11	*Cyanothece*	*Nitzschia* dcl	*Amphora +Seminavis*		
*	*Strobilidium*	*Fabrea salina*	*Entomoneis*		

Table 9: Dominant taxa found at MT from October 2010-September 2011 where dominant taxa are defined as those contributing ≥ 10% of the nano- and microplankton abundance and biomass (*).

Month/Site	Mouth			
Oct-10	*Chroomonas*	*Oxyrrhis marina*		
*	*Oxyrrhis marina*	*Chroomonas*		
Nov-10	*Chroomonas*	*Oxyrrhis marina*		
*	*Alexandrium*	*Oxyrrhis marina*	*Euglena*	
Dec-10	*Chlamydomonas*	*Chroomonas*		
*	*Chroomonas*	*Oxyrrhis marina*	*Chlamydomonas*	*Laboea*
Jan-11	*Chlamydomonas*	*Chroomonas*		
*	*Strombidinopsis*	*Prorocentrum*		
Jan-11	*Chlamydomonas*	*Chroomonas*	*Prorocentrum*	
*	*Chlamydomonas*	*Prorocentrum*	*Chroomonas*	
Feb-11	*Merismopedia*			
*	*Merismopedia*			
Mar-11	*Merismopedia*			
*	*Merismopedia*	*Chroomonas*	*Gymnodinium*	
Apr-11	*Oxyrrhis marina*	*Navicula* b	*Nitzschia* a	*Diplonies* sp 2
*	*Fabrea salina*	*Oxyrrhis marina*	*Gymnodinium*	*Euglena*
May-11	*Chroomonas*			
*	*Chroomonas*	*Strombidinopsis*		
Jun-11	*Chlamydomonas*	*Chroomonas*		
*	*Strombidinopsis*			
Jul-11	*Chlamydomonas*			
*	*Chlamydomonas*	*Codonella*	*Chroomonas*	*Gymnodinium*
Aug-11	*Chlamydomonas*			
*	*Chlamydomonas*	*Gymnodinium*	*Strombidinopsis*	*Protoperidinium*
Sep-11	*Chlamydomonas*			
*	*Protoperidinium*			

Table 10: Dominant taxa found at CC from October 2010-September 2011 where dominant taxa are defined as those contributing ≥ 10% of the nano- and microplankton abundance and biomass (*).

Month/Site	Charters Creek				
Oct-10	*Navicula* , nbl	*Gyrosigma*			
*	*Strombidinopsis*	*Gyrosigma*			
Nov-10	*Gyrosigma*	*Auliscus+Actinoptichus*			
*	*Actinoptichus*	*Gyrosigma*			
Dec-10	*Nitzschia longissima*	*Gyrosigma*			
*	*Gyrosigma*	*Actinoptichus*			
Jan-11	*Oscilatoria*	*Nitzschia* spp.			
*	*Oscilatoria*	*Strombidinopsis*	*Auliscus+Actinoptichus*		
Jan-11	*Lyngbya*	*Chlamydomonas*	*Nitzschia longissima*	*Amphora*	
*	*Lyngbya*	*Strombidinopsis*			
Feb-11	*Nitzschia longissima*				
*	*Strombidinopsis*	*Nitzschia longissima*	*Condylostoma*		
Mar-11	*Chroomonas*	*Cerataulina*	*Chlamydomonas*		
*	*Cerataulina*	*Gymnodinium*	*Chlamydomonas*		
Apr-11	*Gyrosigma*				
*	*Gyrosigma*	*Auliscus/Actinoptichus*			
May-11	*Chlamydomonas*	*Chroomonas*	*Gyrosigma*		
*	*Gyrosigma*	*Chroomonas*	*Chlamydomonas*		
Jun-11	Other *Nitzschia* spp.	*Gyrosigma*	*Nitzschia* , U2	*Entomonies* sp3	
*	*Gyrosigma*	*Auliscus+Actinoptichus*	*Gymnodinium*		
Jul-11	*Chlamydomonas*	*Chroomonas*			
*	*Chlamydomonas*	*Chroomonas*			
Aug-11	*Cerataulina*	*Diplonies* sp 1	*Diplonie* s sp 2	Melosiraceae	*Gyrosigma*
*	*Cerataulina*	*Diplonies* sp 1	*Strombidinopsis*		
Sep-11	*Tryblionella*	*Diplonies* sp 1	Heliopeltaceae		
*	*Actinoptichus*	*Diplonies* sp 1	*Auliscus*	*Tryblionella*	

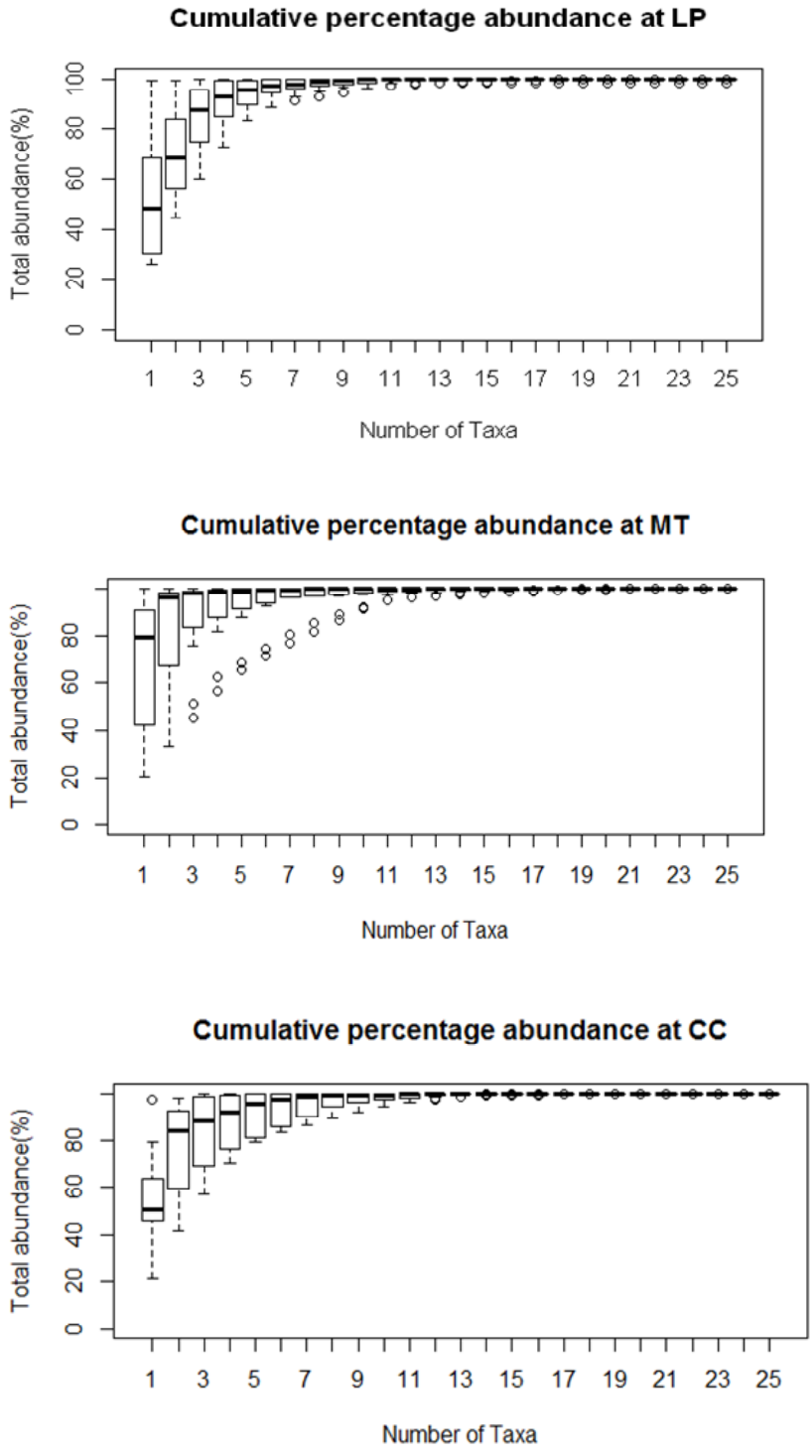

Figure 7: Cumulative percentage abundance of nano-microplankton in LP, MT and CC with respect to number of taxa.

Table 11: Dominant taxa recorded with salinity ranges within which they occurred during this study and in previous St. Lucia studies. ND = no data available (taxa not listed before by previous studies on St. Lucia) and * represents data from benthic samples.

Taxa	Salinity range in this study	Salinity range of other St. Lucia studies	Reference
Cyanothece	47.9-157.32	40-220	Muir &Perissinotto, 2011
Lyngbya	9.00	7.6-29*	Millard & Broekhuysen 1970
Merismopedia	6.92-10.33	ND	
Oscillatoria	17.03	ND	
Chlamydomonas	4.62-57.76	100	Johnson, 1977
Chroomonas	4.93-22.44	ND	
Oxyrrhis marina	12.7-57.76	ND	
Prorocentrum	4.93	42-48	Grindley& Heydorn 1970
Amphora	9.00-50.2	10-24	Bate &Smailes, 2008
Actinoptychus	14.08	6-26	Johnson, 1977
Auliscus	51.38	ND	
Cerataulina	8.55-13.79	ND	
Diploneis	8.55-14.08	14-44	Bate &Smailes, 2008; Johnson, 1977 and Cholnoky, 1968
Entomoneis	14.51	66*	Bate &Smailes, 2008
Gyrosigma	8.55-63.65	4-92*	Bate &Smailes, 2008
Melosira	8.55	8-45	Johnson, 1977; Cholnoky, 1968
Navicula	12.17-69.32	ND	
Nitzschia	12.17-69.37	2-50	Johnson, 1970
N. longissima	9.00-63.65	12-50	Johnson, 1970
N. sigma	45.23-47.9	16*	Bate &Smailes, 2008; Cholnoky, 1968
Tryblionella	14.08	ND	

3.3 Nano- and microplankton biovolume and biomass

3.3.1 Biovolume and biomass

There were no significant differences in the nano and microplankton biovolume (F= 1.2282, p= 0.292) and biomass (F= 1.2617, p= 0.29) between seasons. However, there were significant differences in the biovolume (F= 6.1825, p<0.001) and biomass (F= 6.2913, p<0.001) between sites (Table 7). Figure 8 shows the separation of the sites with respect to abundance, biovolume and biomass. The ranges of estimated biovolume and biomass values of the 5 main groups have been recorded in table 12.

At LP, the highest biovolume was recorded for cyanobacteria ($4.8x10^{10}$µm³/L) followed by diatoms ($3.1x10^{9}$µm³/L). Although ciliates had the lowest abundance at this site, the lowest biovolume of$2.2x10^{8}$µm³/L was recorded for green algae (Figure 9). October to December 2010, cyanobacteria accounted for >95% in total biovolume and >85% in total biomass of the nano- and microplankton

community in LP with ciliates accounting for <5% in biovolume and <10% in biomass. Diatom biovolume dominated with >50% from January 2011 until the end of the study in September 2011 except in April 2011 when cyanobacteria accounted for >60% of the biovolume. Throughout this study, green algae were present but never more than 20% of total biovolume and biomass (Figure 10). However, ciliates and cyanobacteria accounted for most of the biomass from January 2011 until the end of the study with ciliates leading in all months except for February, April and August 2011 when cyanobacteria accounted for 50% of the biomass.

At MT, a peak in cyanobacteria biovolume (4.3x109 $\mu m^3/L$) was recorded in February 2011 due to *Merismopodia* sp. Cyanobacteria had the highest mean biovolume of $3.5x10^8$ $\mu m^3/L$ while green algae had the lowest ($1.13x10^8$ $\mu m^3/L$). However, the lowest biovolume ($2.94x10^3$ $\mu m^3/L$) recorded per sampling session was by cyanobacteria in summer month of October 2010. Diatoms reached their peak biovolume of $1.23x10^9 \mu m^3/L$ in the summer month of January 2011 (Figure 9). In Late 2010 (October and November), dinoflagellates had the highest (>70%) biovolume and biomass followed by green algae (>20% and >15% biovolume and biomass, respectively) in MT. Dinoflagellate biovolume and biomass later increased again at the end of the study period (August and September 2011). Diatoms flourished as their biovolume increased from <1% in October 2010 to >80% in January 2011. However, diatom biomass was below 25% throughout the study except for September 2011 when diatoms recorded a biomass of above 85%. Green algae were present at all sampling sessions with no particular trend, accounting for less than 1% to 60% of biovolume and biomass. With an exception of April and September 2011, cryptophytes accounted for 1%-60% of the biovolume and biomass. Ciliates accounted for less than 30% of the biovolume in all sampling sessions except in July 2011 when they had a percentage biovolume of >75%. Ciliate biomass on the other hand also accounted for less than 30% with exceptions of January, May and June when >45% was recorded. Generally, cyanobacteria accounted for the least (<10%) percentage biovolume and biomass except for February 2011 when >90% were recorded (Figure 10).

At CC, no noticeable trend was found. Diatoms had the highest biovolume in all sampling sessions except for late January 2011 when that of cyanobacteria was higher ($5.97x10^8$ $\mu m^3/L$). Diatoms thus had the highest mean biovolume ($4.77x10^8$ $\mu m^3/L$) while dinoflagellates had the lowest ($2.31x10^6 \mu m^3/L$). Although dinoflagellates had the lowest mean biovolume, the lowest biovolume recorded per sampling session was by green algae in spring months of November 2010 and September 2011 (Figure 9). Diatom biomass also dominated (>60%) throughout the study except for October 2010 when ciliates dominated (>80%), in January 2011 when cyanobacteria dominated (>60%), in April 2011 when dinoflagellates

dominated (>70%) and lastly in July when green algae accounted for more than 70% of the biomass. Dinoflagellates had the lowest biovolume of <2% throughout the study (Figure 10).

Table 12 Biovolume in $\mu m^3/L$ and biomass in pg/L (*) of main nano- and microplankton groups in the three sampling sites.

Main group/Site	LP	MT	CC
Diatom	$3.29x10^6$-$1.98x10^{10}$	$3.43x10^5$-$1.23x10^9$	$3.77x10^7$-$2.22x10^9$
*	$7.02x10^4$-$8.71x10^7$	$1.54x10^2$-$1.32x10^7$	$5.77x10^5$-$1.45x10^7$
Cyanobacteria	$1.01x10^7$-$3.08x10^{11}$	$2.94x10^3$-$4.03x10^9$	$1.75x10^3$-$5.97x10^8$
*	$9.02x10^5$-$1.46x10^{10}$	$7.18x10^2$-$2.63x10^8$	$2.63x10^2$-$4.13x10^7$
Green algae	$2.09x10^2$-$1.76x10^9$	$4.46x10^4$-$3.25x10^8$	$6.92x10^2$-$4.75x10^8$
*	$3.22x10^2$-$1.19x10^8$	$5.50x10^3$-$2.38x10^7$	2.74x10-$3.33x10^7$
Cryptophytes	$13.8x10^4$-$9.82x10^6$	$2.75x10^3$-$1.80x10^8$	$8.02x10^2$-$1.38x10^8$
*	$1.82x10^3$-$8.72x10^5$	$4.02x10^2$-$1.34x10^7$	$1.27*10^2$-$1.04x10^7$
Dinoflagellate	$4.42x10^3$-$1.01x10^9$	$1.40x10^5$-$1.04x10^9$	$7.11x10^3$-$2.41x10^7$
*	$1.94x10^3$-$6.83x10^7$	$5.10x10^4$-$1.17x10^9$	$9.81x10^2$-$4.78x10^6$
Ciliate	$1.61x10^7$-$1.04x10^{10}$	$6.86x10^3$-$1.17x10^9$	$2.54x10^4$-$5.53x10^7$
*	$3.06x10^6$-$1.97x10^9$	$8.62x10^3$-$2.21x10^8$	$5.04x10^2$-$1.05x10^7$

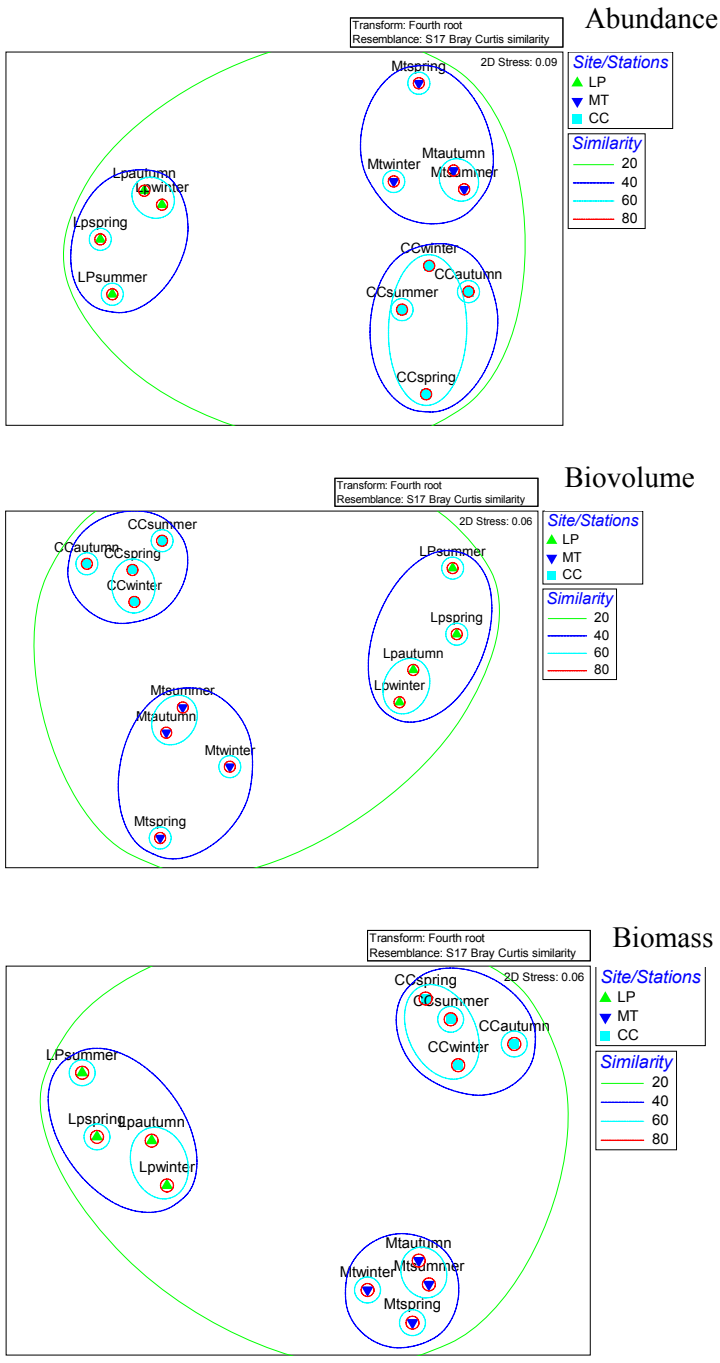

Figure 8: Multidimensional Scaling (MDS) of community assemblage based on abundance and biovolume.

With respect to groups and sites, the highest total biovolume and biomass for all groups was recorded in LP except for green algae with its maximum total biovolume in MT. CC had the least total biovolume and biomass for all five groups except for diatoms whose least total biovolume was recorded in MT and dinoflagellates which had the least total biomass in MT.

Ciliate biomass and biovolume was highest in LP ($3.25x10^8$ pg/L and $1.89x10^9$ µm³/L respectively) accounted for by the presence of *Fabrea salina* and hypotrichs of the subclass Hypotrichia and genus *Euplotes*. The mouth had the second highest biomass and biovolume of ciliates ($2.38x10^7$ pg/L and $1.25x10^8$ µm³/L respectively) accounted for by the presence of Tintinnids and Choreotrichs of the subclass Choreotrichia and least ciliate biomass and biovolume was recorded in CC ($2.30x10^6$pg/L and $1.22x10^7$ µm³/L).

There was a significant correlation (R= 0.547, $p<0.001$) between biomass (carbon) from biovolume measurements and biomass (carbon) calculated from chl a measurements using a fixed conversion factor of 1:50 for chl a:C. There were however, significant differences between the means of both biomasses ($p<0.001$. F = 15.341) falling within the range $4.6x10^7$–$2.13x10^9$ pg/L for biomass from chl a and $2.23x10^6$–$1.46x10^{10}$ pg/L for biomass from biovolume. Chlorophyll a:C ratio was highly variable with time and differed between sites with the highest ratio in CC (0.06-4.31), the lowest in LP (0.002-1.06) and an intermediate range at MT (0.01-2.86) (Figure 11). It should be noted that the chl a:C ratio was at times higher than the known 0.02 stated in Reynolds (2006). Therefore, there was more cellular carbon in LP relative to chl a and the reverse was true for CC. There was however, a strong and significant positive correlation (r= 0.652, $p<0.001$, n=36) between chl a and autotrophic carbon biomass.

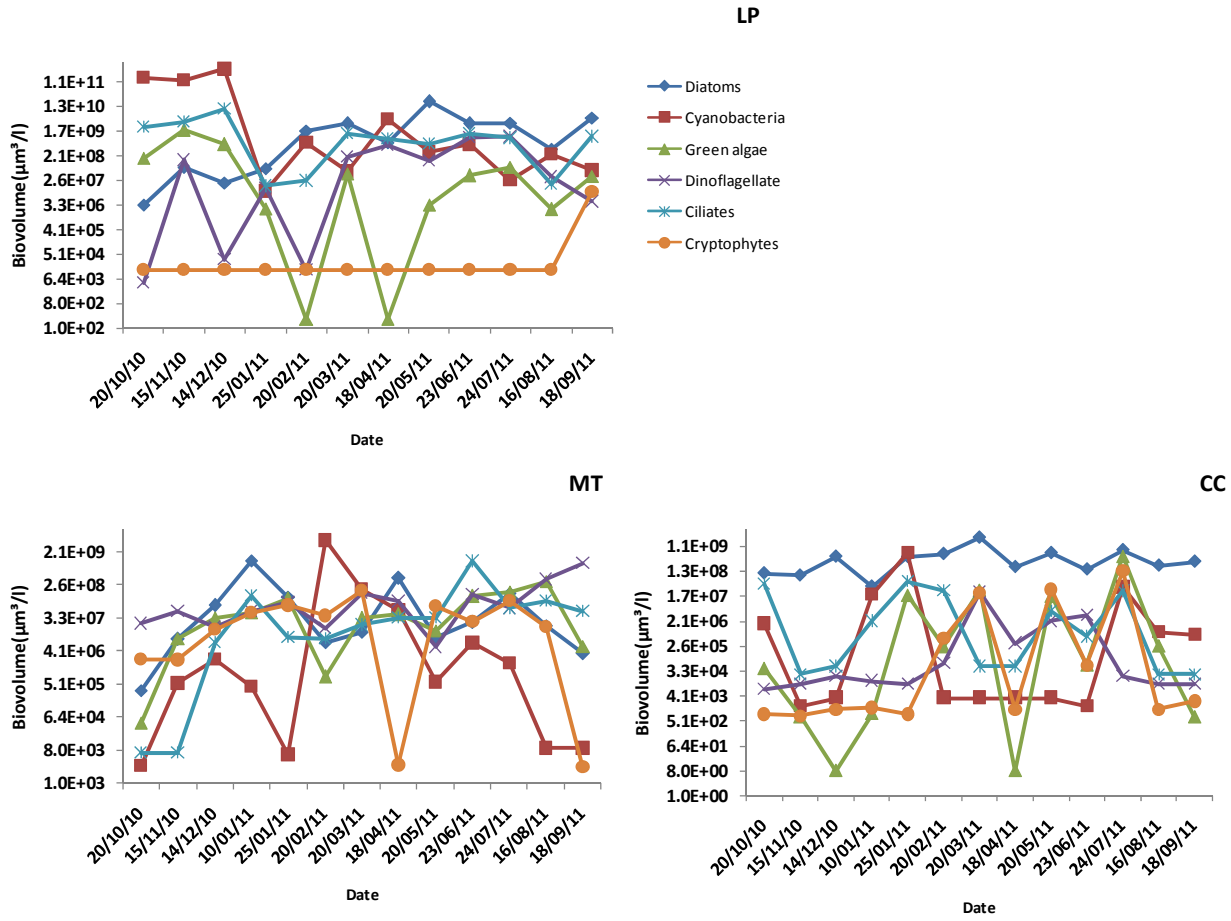

Figure 9: Mean biovolume (µm³/L) of the main nano- and microplankton groups in LP, MT and CC.

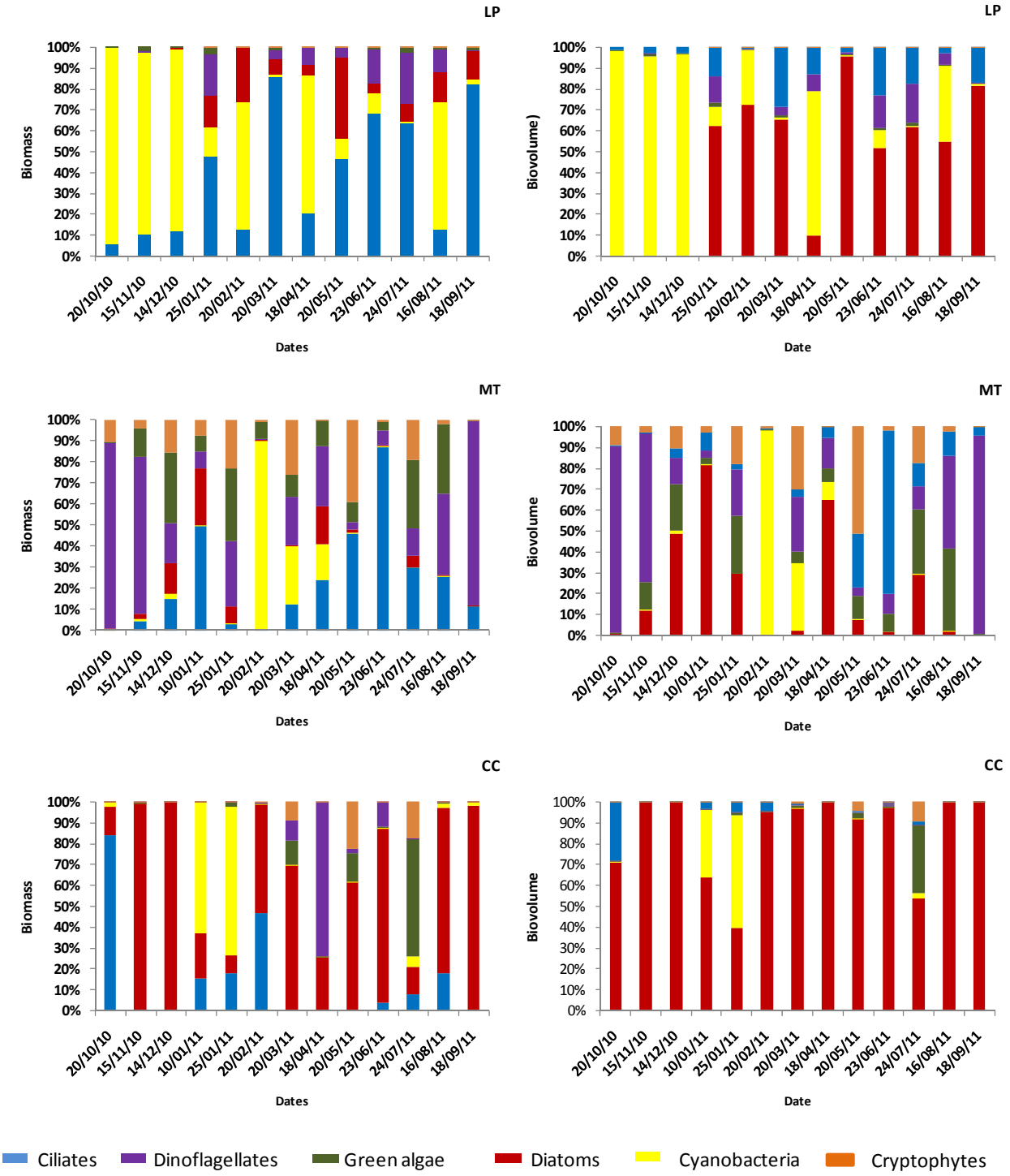

Figure 10: Percentage biovolume and carbon biomass of the main nano- and microplankton groups at LP, MT and CC from October 2010-September 2011.

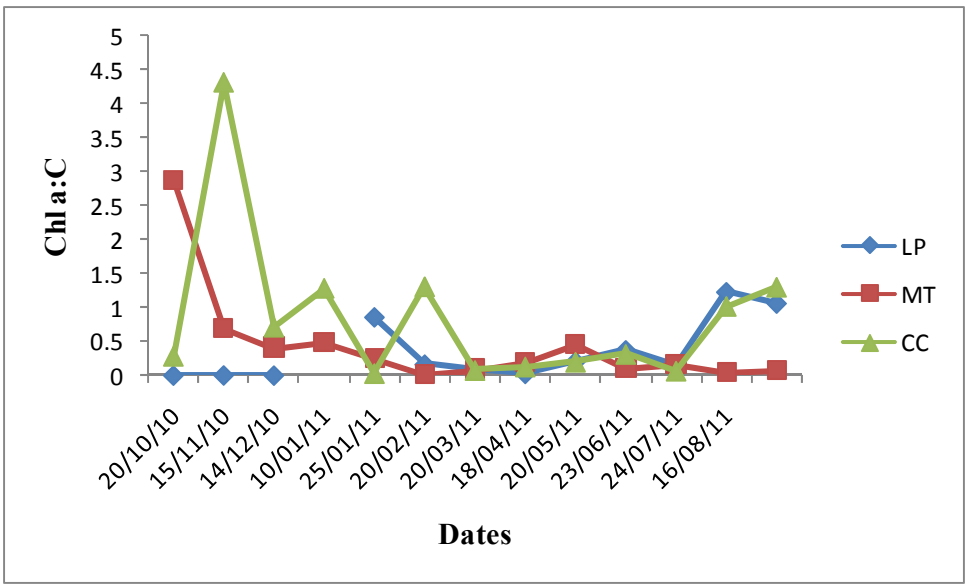

Figure 11: Changes in chlorophyll a:carbon ratio per date at all 3 sites.

3.4 Autotrophy and heterotrophy

3.4.1 Abundance

Throughout this study, more than 80% of the nano- and microplankton community in LP was made up of autotrophs in terms of abundance. At MT, with the exception of October and November 2010 when heterotrophic biovolume was between 40-28%, autotrophs accounted for more that 85% of the total abundance throughout this study. With the exception of October 2010 when heterotrophic biovolume was 15%, autotrophs accounted for more than 99% of the total abundance in CC throughout this study (Figure12). The average autotrophic abundance per site in a descending order was $2.04x10^8$ Cells/L at LP, $1.75x10^6$ Cells/L at MT and$4.89x10^5$ Cells/L at CC. The average heterotrophic abundance per site in descending order was $2.14x10^5$ Cells/L at CC, $1.84x10^4$ Cells/L at MT and $1.11x10^3$ Cells/L at LP. Generally, the nano- and microplankton in this system was thus dominated by autotrophic species with CC having the highest abundance of heterotrophs.

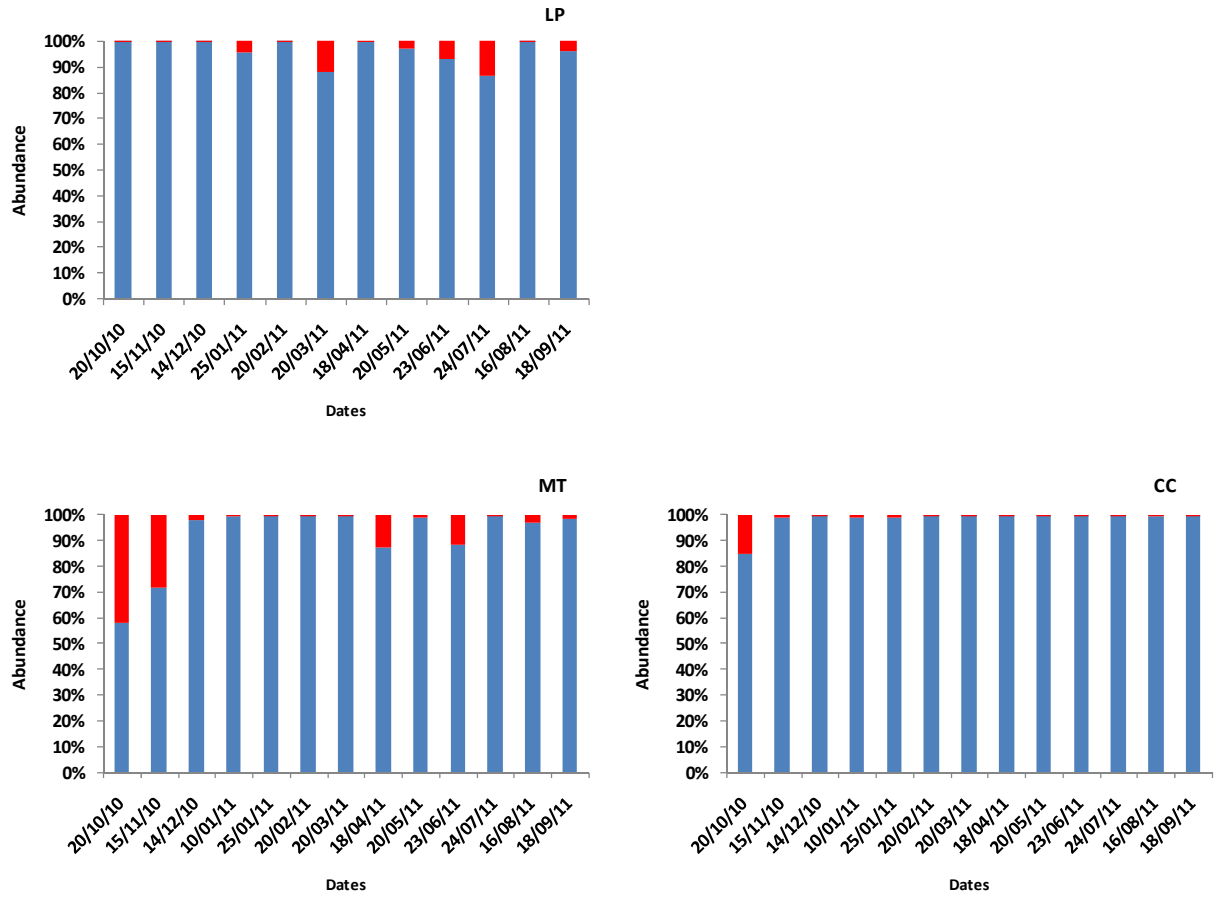

Figure 12: Percentage abundance of autotrophs (blue) versus heterotrophs (red) in LP, MT and CC.

3.4.2 Biovolume and biomass

In LP, 98% of the biovolume of nano- and microplankton in October to December 2010 was made of autotrophs. During this period also, autotrophs also contributed >88% of the biomass. Thereafter, autotrophs dominated (>60%) in biovolume throughout the study except for August 2011 when they only accounted for 40% of the total biovolume in LP (Figure 13). The dinoflagellates of the genus *Protoperidinium*, *Oxyrrhis*, and ciliate genus *Fabrea* accounted for the increase in heterotrophic biovolume in August 2011. Heterotrophic biomass on the other hand was above 50% from January 2011until the end of the study with an exception of February, April and September 2011 when autotrophs accounted for more than 50% of the biomass and in May 2011 when the ratio of autotrophic versus heterotrophic biomass was 1.

Similarly, throughout the study in MT, >70% and >50% of the total biovolume and biomass respectively, was made of autotrophs except for October 2010 and June 2011 when heterotrophs accounted for more than 80% of the total nano- and microplankton biovolume(Figure 13). The heterotrophic dinoflagellate of

the genus *Oxyrrhis* was responsible for the increase of heterotrophic biovolume in both months. However, in CC, autotrophic biovolume was >95% throughout the study except in October 2010 which had a heterotrophic biovolume of>25% (Figure 13) caused by the ciliates of the subclasses Choreotrichia and Oligotrichia. Biomass of autotrophs was also above 80% throughout this study except for October 2010 when it dropped to 15% and 52% in February. With respect to the nano- and microplankton of this system, most of the energy available in terms of carbon (biomass) is provided by autotrophs. The mean autotrophic biovolume per site in a descending order was LP ($5.18x10^{10}$ $\mu m^3/L$), CC ($5.85x10^8 \mu m^3/L$) and MT ($5.18x10^8 \mu m^3/L$). Whereas the autotrophic biomass was least in CC ($1.29x10^7$ $\mu m^3/L$), followed by MT ($4.43x10^7$ $\mu m^3/L$) and highest in LP ($2.41x10^9$ $\mu m^3/L$).

Overall, ciliates contributed more to the heterotrophic biomass (HB) in the system than heterotrophic dinoflagellates (HDF). Ciliates biomass contributed more than 50% of HB throughout the system except in August 2011 at LP, October-December 2010 in MT and November 2011 in CC (Figure 14).

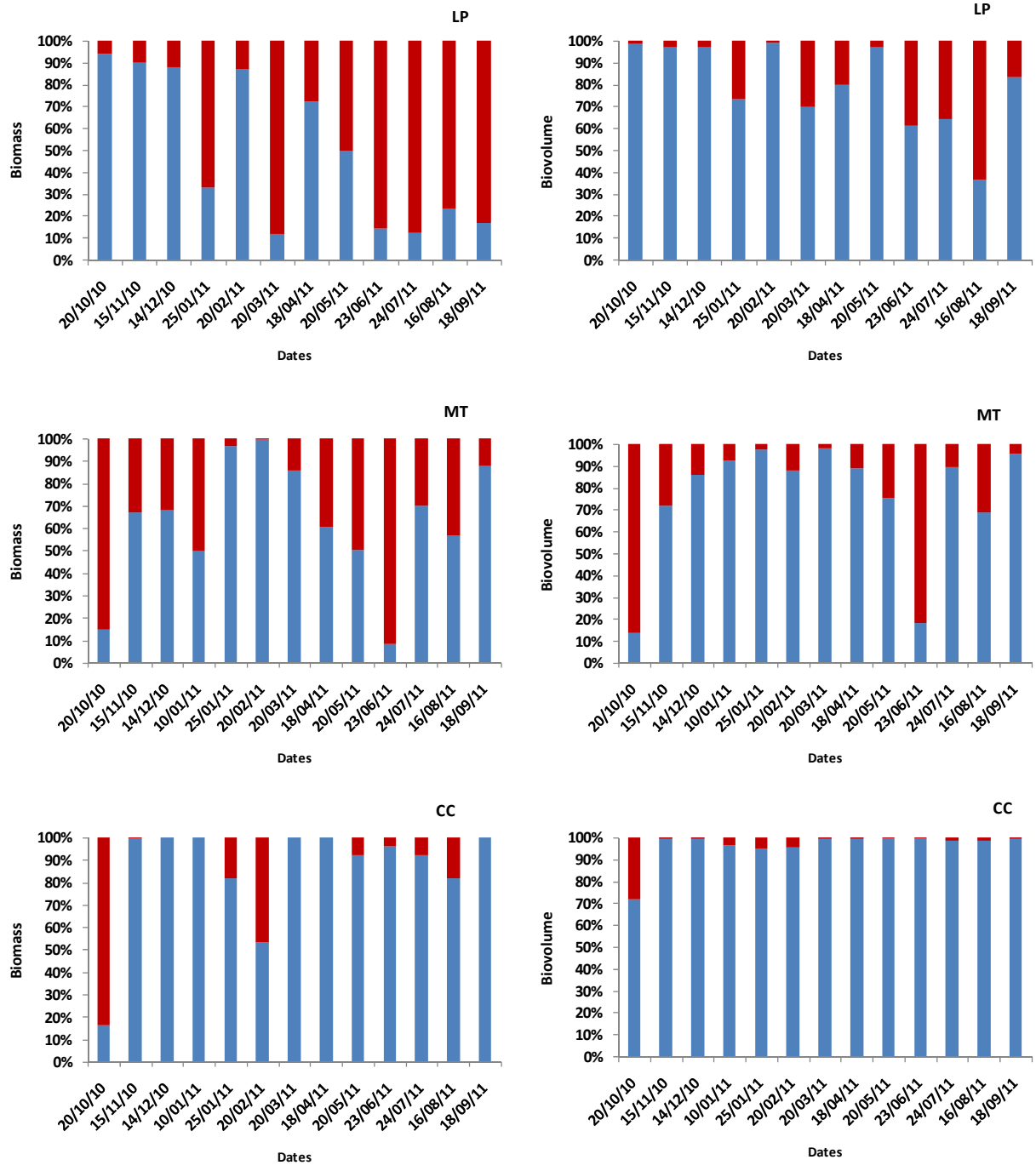

Figure 13: Percentage biovolume and biomass (Carbon) of autotrophs (blue) verses heterotrophs (red) in LP, MT and CC per all sampling sessions.

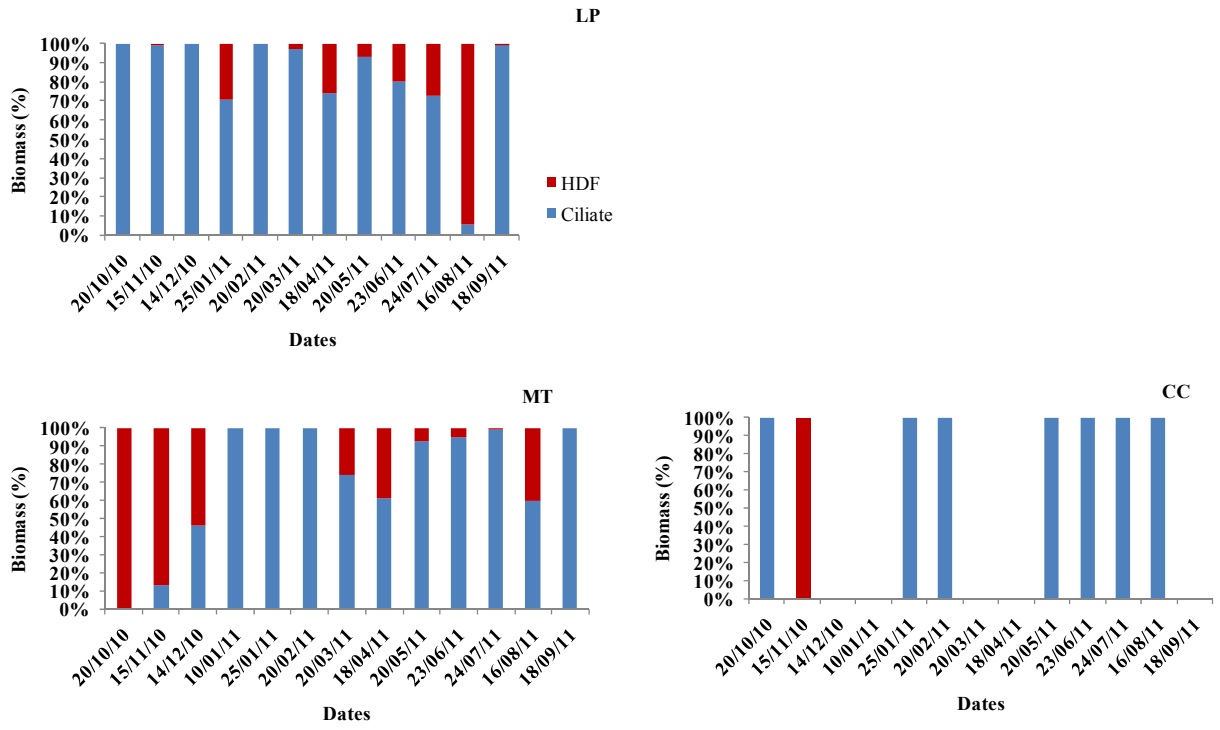

Figure 14: Percentage contribution of ciliates and heterotrophic dinoflagellates (HDF) to the heterotrophic biomass in the system per date for all 3 sites. The empty spaces in CC are dates when no heterotrophs were recorded.

3.5: Effects of environmental variables on community structure.

Keeping every other variable constant, salinity accounted for 44.3% of the variability in the abundance of nano- and microplankton (Table 13) and had a significant and strong correlation (p= 0.003, R= 0.561) with abundance also (Table 14). The depth of the system and the light attenuation coefficient (Kd) had a weak but significant correlation (p= 0.037, R= 0.388; p= 0.034, R= 0.395 respectively) with abundance. Other variables which were seasons, turbidity, Nitrogen:Phosphorus (N:P) ratio and temperature did not show a significant correlation with the nano- and microplankton abundance in the system during this study (Table 13). A one-sample Kolmogorovs-Smirnov test for normality performed on the studentized residuals proved that data was normally distributed (p= 0.902).

Taxon richness on the other hand, demonstrated a weak but significant negative correlation (R= -0.392, p= 0.035) with temperature. No other variable had a significant relationship with taxon richness. However, diversity index was significantly negatively correlated with the site and depth (p= 0.012, R= 0.479 and p= 0.006, R= -0.746, respectively) of the system. There was a very strong (R= 0.909) and significant (p<0.001) correlation between biomass and abundance of the nano- and microplankton. There

was thus a strong positive correlation between abundance, biovolume and biomass of these organisms (Table 14).

The variance in biomass however was accounted for by the diversity index, salinity, Kd and depth (Table 14). With every other variable kept constant, salinity accounted for 52.8% of the variability in biomass. Kd and depth only accounted significantly for the variability in biomass when salinity was excluded (Table 11) Biovolume on the other hand is influenced by salinity (p= 0.002, R= 0.587), Kd (p= 0.003, R= 0.572) and N:P (p= 0.029, R= -0.410). Salinity accounts for 34.4% of the variability in biovolume if every other variable was kept constant. Taxon richness and N:P also account for the variance in biovolume (Table 13). Overall, salinity was the main factor responsible for the variation in abundance, biovolume and biomass of the phytoplankton during this study (Table 13).

Table 13: Coefficient of determination (R^2), sum of squares and variables derived from a multiple linear regression analysis.

R squared	Sum of squares	Dependent variable	Model/Independent variables
.443	23.705	Abundance	Salinity
.547	29.247	Abundance	Salinity and turbidity
.677	36.267	Abundance	Salinity, turbidity and taxon richness
.344	5.072	Biovolume	Salinity
.604	8.899	Biovolume	Salinity and taxon richness
.689	10.144	Biovolume	Salinity, taxon richness and N:P
.528	16.949	Biomass	Salinity
.269	5.305	Biomass	Kd
.486	9.587	Biomass	Kd and depth

Table 14: Variables, their significant Pearson correlations and *p* values (*significant at 0.01< p ≤ 0.05, **significant at 0.001 < p ≤ 0.01, ***significant at p ≤ 0.001) based on Multiple Linear Regression Analysis. Data points unmarked are not significant i.e. *p*> 0.05.

	Abundance	Biomass	Biovolume	Taxon richness	Diversity index	Turbidity	Salinity	Kd	Temperature	Depth
Abundance	1.000	.909***	.869***	.242	.185	-.048	.561**	.395*	.001	.388*
Biomass		1.000	.955***	.309	.373*	.083	.609***	.519**	.004	.432*
Biovolume			1.000	.291	.280	.076	.587**	.527**	.018	.316
Taxon richness				1.000	.153	.089	-.326	.013	-.392*	.287
Diversity index					1.000	.269	.306	.094	-.077	.525**
Turbidity						1.000	.266	.498**	-.339	.060
Salinity							1.000	.629***	.016	.263
Kd								1.000	-.017	-.064
Temperature									1.000	-.130
Depth										1.000

According to a CCA, The assemblage of dominant taxa was strongly associated with physico-chemical variables (p<0.01, pseudo F= 3.60). This CCA explains only 36.01% of the variation in the abundance and occurrence of dominant abundant taxa. The two axes (F1 and F2) accounted for 85.17% of the variation in the occurrence and abundance of the abundant taxa during this study (Table 15). The main physico-chemical variables in F1 were depth and salinity while the main physico-chemical variables in F2 were temperature and salinity (Table 16). From the CCA plot in Figure 15, the cyanobacterium *Merismopedia* and the dinoflagellate *Prorocentrum* increase with an increase in depth and a decrease in turbidity. The cryptophyte *Chroomonas*, the chlorophyte *Chlamydomonas* and diatoms *Diploneis*, *Nitzschia longissima* dominate with a decrease in salinity while *Oscillatoria* and *Cyanothece* increase with an increase in salinity. Nitrogen:Phosphorus ratio had a weak correlation with all taxa. Although these taxa occur and survive in the high salinities of this system, these salinities are not ambient for their growth.

Table 15: Eigenvalues (CCA) showing cumulative strengths of the axis.

	CCA1	CCA2	CCA3	CCA4	CCA5
EigenValue	0.778	0.413	0.157	0.033	0.018
Proportion explained	0.556	0.295	0.112	0.023	0.013
Cumulative proportion	.0556	0.852	0.964	0.987	1.000

Table 16: Principle coordinates values of the variables in the CCA plot.

	CCA1	CCA2	CCA3	CCA4	CCA5
Turbidity	-0.028	-0.333	-0.379	0.614	-0.558
Salinity	-0.843	0.061	0.073	0.445	-0.229
N:P	0.068	0.044	0.308	0.984	0.0189
Temperature	-0.451	0.413	0.539	-0.259	-0.464
Depth	0.924	0.474	-0.093	0.121	0.002

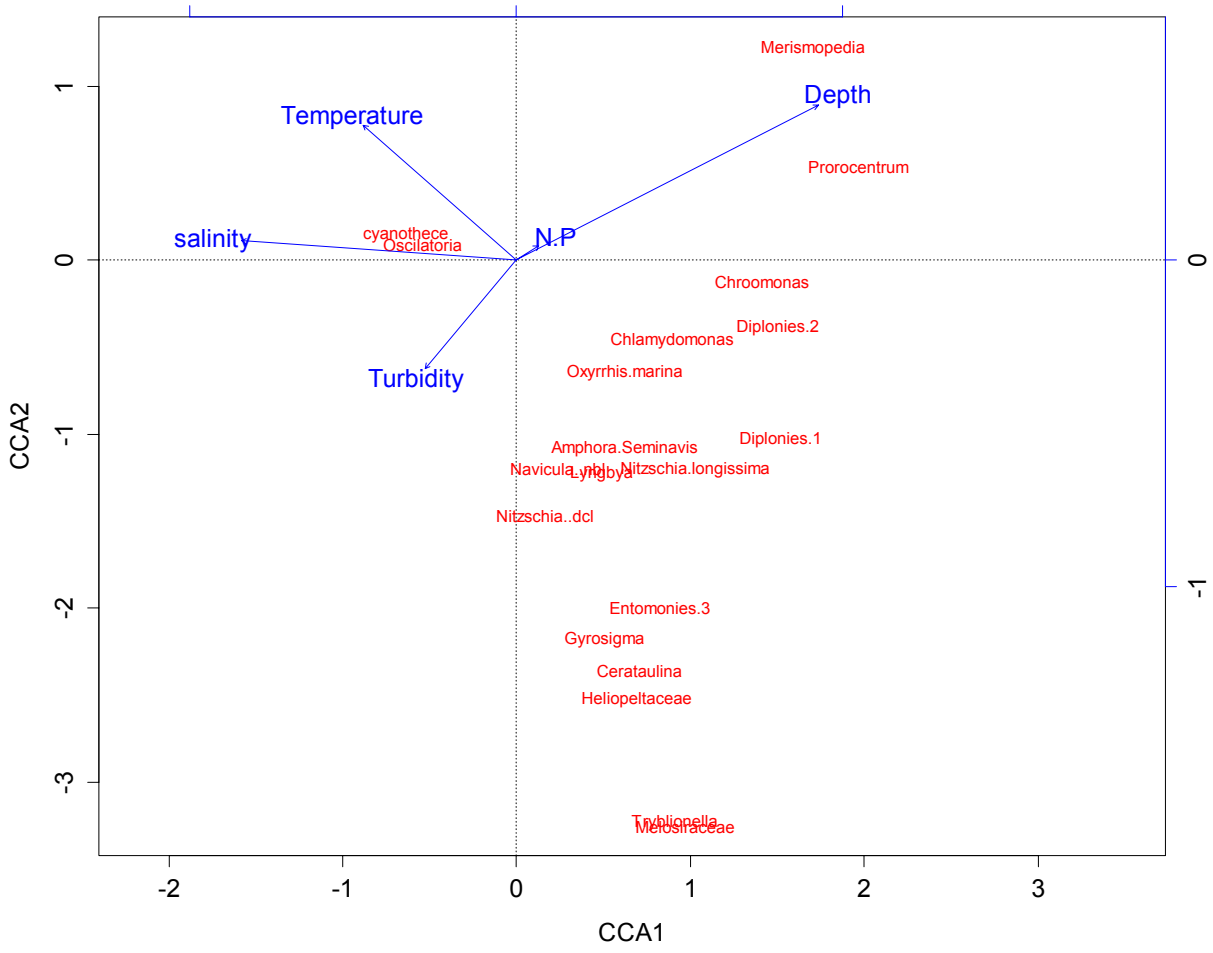

Figure 15: Canonical Correspondence plot relating abundance of dominant taxa and physico-chemical variables.

CHAPTER 4

DISCUSSION

The aim of this study was to characterize the nano- and microplankton (autotrophic and heterotrophic) at different sites of the lakes and investigate their temporal changes under the influence of changing physico-chemical conditions. The changes in the nano and microplankton community composition, abundance, biovolume and biomass, were analysed. There was no difference in the species composition, biovolume, abundance and biomass between seasons but there was high variability over time from month to month. However, there were significant differences between sites. Overall, in the northern embayment cyanobacteria dominated in terms of abundance, biovolume and biomass (biological variables) with the main taxa being *Cyanothece* (Table 8). In the Channel, green algae and cryptophytes dominated in all three biological variables with the *Chlamydomonas* and *Chroomonas* as the main taxa (Table 9). In South Lake, green algae and cryptophytes dominated in terms of abundance. Diatoms with main taxa Triceratiaceae, *Gyrosigma*, *Diploneis* and *Nitzschia* (Table 10) dominated in biovolume and biomass. Although other factors such as nutrients, temperature, and light intensity are responsible for variations in plankton community composition and biomass, in this study, salinity was the main environmental variable responsible for this variation between sites.

4.1 Physico-chemical variables

Salinities in the northern region were the highest in the system. This is similar to previous studies of the system (Cyrus et al., 2010; Carrasco & Perissinotto, 2012). This high salinity was due to high evaporation rates and limited freshwater input to the system. The high salinities as was the case of this study have always been noted to decrease with the onset of the rainy season (Bate & Smailes 2008; Tirok & Scharler 2013). The N:P ratio was highest near the mouth when compared to other sites of the system. This could be because the Channels received some freshwater though limited from the Mfolozi river. Also, during the open phase from February to August 2007, dissolved inorganic nitrogen and dissolved inorganic phosphorus was lower compared to what was recorded in the closed phase of 2004-2005 (Perissinottoet al., 2010). This would suggest that nutrients decrease and are possibly limiting during the open mouth phase whereas they increase in the closed phase which was the case of the present study. Johnson (1977) concluded that the system (open mouth state) in 1975 was oligotrophic. Ammonia values recorded here are within the range of those recorded in the Sundays estuary suggested to be nutrient rich in nature (Kotsedi et al., 2012). The system at time of sampling was thus not nutrient deficient as also pointed out by Tirok & Scharler (2013) and phosphorus was always available in excess when compared to nitrogen.

In estuarine systems, freshwater is one of the main sources of nutrients. The system however had limited freshwater supply during the droughts and its main source of extra nutrients was defecation and foraging

of animals such as the hippopotamus as they stirred up nutrients in the sediments also (Perissinotto et al., 2013). This coupled with the presence of cyanobacteria in the system as another source of nitrogen (Perissinotto et al., 2013) would explain why the system was not limited in nutrients although it was limited in freshwater.

4.2 Nano- and microplankton community composition

The non- significant difference between seasons was a result of the monthly variability experienced which masked the overall changes per season. This is similar to what was suggested by Tirok & Scharler (2013) who also recorded no significant differences in chl a (a proxy for biomass) between seasons. Johnson (1977) also reported that there was no seasonal difference in the community composition of phytoplankton in the system. Johnsons' (1977) study was conducted when the mouth was open and this study when the mouth was closed but there was still no difference between seasons suggesting that the state of the mouth only affects the type of species found in the system but not the succession or changes in the community composition over a short (1-3 years) time scale. Also, the absence of seasonal patterns in nano- and microplankton community composition could be because rainfall which controls salinity, the main physico-chemical variable in the system is erratic in South Africa. Hence changes in community structure are opportunistic responding to episodic freshwater discharges (Hilmer, 1990; Allanson & winter, 1999). These results are in sharp contrast to estuarine lakes and bays in temperate regions that experience cold dark winters and warm light summers. In St. Lucia, no strong seasonal variations were experienced with all seasons being relatively warm (>18□ C) with similar day length. Seasonality (temperature and light)in temperate regions thus explains much of the variance in the community composition as there is a clear succession pattern between the seasons with diatoms dominating in winter and non-diatom dominating during summer (Adolf et al., 2006). Although there was no difference between seasons, there were significant differences in community composition, and the biological variables between sites.

The main taxon in the northern embayment, *Cyanothece* is saline tolerant and flourishes in systems with hypersalinity (Roussomoustakaki & Anagnostidis, 1991; De Phillipis et al., 1993; Margheri et al., 1999;Welsh et al., 2008) and high temperatures. The *Cyanothece* bloom was first recorded in 2009 when salinities of above 220 were recorded in False Bay (Muir & Perissinotto, 2011). In this study also, this taxon was only recorded in the northern embayments and absent in the Channel and southern lake that had lower salinities. This taxon reduced in abundance with the onset of rains in February 2011 (Muir & Perissinotto 2011) further proving its affinity to higher saline environments. The taxon *Synechococcus bacillaris* of the same family Synechococcaceae as *Cyanothece* which is commonly misidentified as *Cyanothece* had the highest abundances in northern basins in 1973 between February and September

when the system was hypersaline but also disappeared in October when salinities were reduced (Johnson, 1977). The dominant diatom taxa that flourished when salinities dropped were *Nitzschia*, *Navicula* and *Amphora* known to be euryhaline in nature and either benthic or pelagic (Round et al., 1990). *Nitzschia longissima* was present throughout the system, dominant in the northern embayment and South Lake as was also recorded by Johnson (1977). High cell counts were recorded at salinities of 50 and 12. The species is able of considerable growth in a wide range of salinities (Johnson, 1977) and thus can be a dominant taxa at high and low salinities. It was suggested that this species in the system tends to be more common in summer (Johnson, 1977) but this was not true for this study as the species was recorded in all seasons. The marine and halophilic genus *Dunaliella* (Post et al., 1983; Reynolds, 2006) was recorded only in LP during spring and early summer (October 2010-December 2010) when salinities ranged from 94-216 and disappeared when salinities dropped to 46 in January 2011. This species was never recorded anywhere else in the system during this study. This genus declined below detection limit when salinities decreased, implying it could strictly tolerate only environments with very high salinities. This is because, the number of cysts called erythrospores (a resting phase which develops when salinities reduce) that proliferate to chlorospores (the form we observe) is a function of the salt concentration (Oren, 2005).*Dunaliella* has been shown to flourish between salinities of 60-230 with 150 as the maximum salinity for optimal growth (Oren, 2005). This is different from the genus *Chlamydomonas* which can tolerate wide range salinities as it was recorded in all 3 sites. This suggests that other factors apart from salinity account for the presence of this genus.

The lower salinities in MT accounted for the dominance of green algae, cryptophytes and a high diversity of diatoms. The high abundance of the cyanobacteria *Merismopedia* in the months of February and March can also be accounted for by salinity as it is suggested to flourish in lower salinities. The *Merismopedia* abundance $10.88x10^7$Cells/L-$15.86x10^7$Cells/L at salinities of 10.8 in February 2011 reduced to 68.35 x 10^5 Cells/L–$76.49x10^5$ Cells/L when salinity increased to 13.78 in March 2011. This was similar to the Myall lakes system in Australia were $17x10^6$ cells/L were recorded at a salinity of 16 but increased to $26.64x10^6$ Cells/L when salinity decreased to 8(Redden& Rukminasari 2008), implying they cannot tolerate high salinities. The genus *Diploneis,* a marine - brackish water diatom, occurred at all sites of this study confirming the records of Johnson (1977) and Cholnoky (1968) who reported the presence of the genus throughout the system and mostly common in the Channel where salinities were generally marine. Similarly in this study the genus was amongst the dominant taxa in MT and CC where salinities were brackish and not dominant in LP where salinities were much higher (>50). The halotolerant chlorophyte *Scenedesmus* and fresh water chlorophyte *Kirchneriella* only occurred in MT. Their abundance and biomass decreased with increase in salinity as they exhibit increase in respiratory rates and decrease

photosynthetic rates when salinities increases (Flameling& Kromkamp, 1994). These two species were also recorded in the shallow (1m average) lake Manzala in Egypt with a maximum salinity of 20.3 (Ramdani et al., 2009).

Green algae especially *Chlamydomonas*, has been recorded in a wide range of salinities (Kotsedi et al., 2012) but flourish most in salinities of 6-52 (Lee et al., 1974). The southern lake of the estuarine system had a lower salinity range of 8.5-66.6 reasons why with respect to abundance, green algae dominated at this site. Of the three sites, only CC had centric diatoms (4 taxa) as dominant species. These centrics have been reported to flourish in waters with low salinities, e.g., the taxon *Melosira* is a fresh water and marine epibenthic taxon (Round et al., 1990) which was first recorded by Johnson (1977) in the system occurring when salinities ranged from 12-16. This study confirms the observation by Johnson (1977) that the genus is most successful in brackish waters in spite of its occurrence in marine waters in St. Lucia. *Navicula*, *Nitzschia*, *Entomoneis* and *Amphora* which were recorded at all sites have been noted before in the studies of eight South African estuaries (Berg, Palmiet, Goukou, Gourists, Great Brak, Keurbooms, Gamtoos and Sundays) to tolerate a wide range of salinities from fresh to marine (Adams & Bate, 1999). The common genera recorded in all eight estuaries was *Amphora*, *Navicula* and *Nitzschia* (Adams & Bate, 1999), which were part of the dominant species in all 3 sites of this study except for MT which did not have *Amphora* as a dominant species. The rest of the dominant diatoms and overall more than 70% of the diatoms (Figure 6) were predominantly brackish and epipelic in nature, hence they were benthic species. Also, only 8 of the 47 taxa listed by Johnson (1977) were truly pelagic and 39 were benthic. These results suggest that resuspension plays an important role in the community composition and biomass of phytoplankton in the water column which was also discussed by Tirok & Scharler (2013). This confirms the suggestion that CC has few true pelagic species and benthic species were resuspended from the sediment by turbulence experienced in the system. Also, Cholnoky (1968) reported the absence of planktonic species in 1963 stating that the shallow and turbulent nature of the system was not suitable for the development of planktonic species (Cholnoky, 1968). The species in the lake Manzala in Egypt, a very turbulent, shallow lake were also mostly of benthic nature (Ramdaniet al., 2009).

Overall, the taxon richness decreased from the mouth region to the northern embayments which concur with Cholnoky (1968) and Johnson (1977). The latter recorded 77 species in the tidal regions; 55 in False Bay and in this study 79 taxa were recorded in the mouth and 56 in LP. This was because fewer species are able to osmoregulate and adapt to hypersaline environments in the northern embayment (Cholnoky, 1968).

Bate & Smailes (2008) who reported on diatoms from 2004-2006 when the mouth was closed and parts of the system were hypersaline, did not record any of the species recorded by Johnson (1977), however, some species recorded by Cholnoky (1968) were recorded in their study. The difference in the state of the mouth was suggested by Bate & Smailes (2008) to explain this difference. From this suggestion, one expects that the diatoms recorded for this study [same conditions as Bate & Smailes (2008)] should be similar to theirs and none of Johnson's taxa should be recorded in this study which however was not true. Hence, although the mouth opening and the influx of sea water in the system has an effect on the phytoplankton community composition, it is not the sole influencing variable.

Overall, as shown by the Canonical Correspondence analysis (CCA) on dominant abundant taxa composition and their abundance, depth, salinity and temperature accounted for most of the variance in the community composition and abundance of the taxa. In South African estuaries, salinity was previously found to be the main factor accounting for the variance in phytoplankton community composition (Adams & Bate, 1999) while in estuaries and lakes in temperate regions, temperature has been the main factor accountable for this variance (Adolf et al., 2006).The concentration of nutrients in the water column were not limiting for algal production in all three sites during period of the study (Tirok & Scharler, 2013). Also, the nutrient concentration did not differ between sites and seasons ($p > 0.05$) hence could not have accounted for a significant difference in the species composition as also shown in the CCA plot (Figure 12). Taxonomic diversity showed no difference with changes in N:P ratio, confirming a suggestion made by Redden & Rukminasari (2008) that enriched treatments of different N:P ratios do not show significant differences, however, with different salinity treatments there were variations in taxonomic diversity. This could be because an increase in salinity had a stronger effect on the type of algal groups than the biomass-limiting nutrients N and P. This situation is unlike that in the Sundays estuary where phytoplankton community structure and blooms are mainly due to variations in nutrients with the upper, middle and lower reaches of the estuary having significantly different nutrient concentrations (Kotsedi et al., 2012).

The changes in phytoplankton community composition have an effect as the phytoplankton species present in a system at a particular time determine to a great extent the species and food availability up the food chain. Primary producers influence directly and indirectly the organisms up in the food chain or higher trophic levels. Heterotrophs basically repackage the parcels of carbon by feeding on bacteria, algae and ciliates (Sherr& Sherr, 1994). Some heterotrophs (zooplankton) are specific in their food source hence feed on a particular algae only while other heterotrophs feed on a wider range of algae. For specific primary grazers, the absence of their food source leads to their extinction from the system at that time.

This would also lead to the extinction of other organisms up the food chain that depend solely on the primary grazer. A typical example showing a linear relationship between algae and other species up the food chain was the "one species – per – level food chain" in LP where the presence of the *Cyanothece* bloom led to the abundance of flamingos in the system (Carrasco & Perissinotto, 2012). Flamingos fed on the copepod *Apocyclops dengizcus* which fed on the ciliate, *Fabrea* (heterotroph) whose main food source was the *Cyanothece* (primary producer) (Carrasco & Perissinotto, 2012).

4.3 Nano- and microplankton biovolume and biomass

Biovolume of groups such as diatoms reached high proportions but comparatively lower biomasses. The presence of large vacuoles in diatoms would account for the high volume but low biomass whereas in cyanobacteria and green algae, smaller biovolume yields higher biomass (Figure 10). Lister's point in False Bay had the highest biomass (Carbon, pg/L) and the lowest was recorded from Charters Creek. Since nutrients and light availability were higher in the Channel and not at LP, the lack of or limited grazers in LP due to its high salinity (Carrasco & Perissinotto, 2012) could be responsible for the high biomass in LP. Grazing can also thus be suggested as the factor that regulated the biomass, reducing biomass in MT and CC which had lower salinities hence a relatively higher abundance of zooplankton (grazers) (Carrasco & Perissinotto, 2012). The appearance of rotifers in LP after the summer rains of January 2011 (Carrasco & Perissinotto, 2012) would also explain the high phytoplankton biomass in LP. This may be because zooplankton such as copepods have been shown to selectively feed on rotifers and other microzooplankton instead of phytoplankton (Yang et al., 2009; Yang et al., 2010; Šorf & Brandl, 2012). The presence of these alternate food sources for grazers would suggest the high phytoplankton biomass. Other than salinity, Kd accounted for 26.9% of the variation in biomass. Although there was no significant difference in Kd between sites, LP had a slightly higher Kd hence received less light while MT had the least Kd hence more light.

The difference in biomass from chl a values and biovolume measurements was due to the difference in methodology. Excluding the three months (Oct-Dec) of 2010 when the cyanobacteria bloom was severe, carbon from biovolume ranged from $2.23x10^{6}$–$3.36x10^{8}$ pg/L only. The reason why biomass ($6.27x10^{9}$-$1.46x10^{10}$ pg/L) from biovolume measurements was higher than biomass ($1.13x10^{9}$-$2.13x10^{9}$ pg/L) from chl a measurements during these three months was because *Cyanothece* the most dominant taxa in LP is known to have a low chl a content (pers. obs. Akash Anandraij).This explains why from Oct-Dec 2010, biomass from biovolume was higher than biomass from chl a. For the rest of the study, biomass from chl a was higher than biomass from biovolume. This was because, during the filtering process for chl a analysis, other photosynthetic organisms such as fragments of macroplankton which were common in the

samples, other microzooplankton with ingested algae were trapped in the filters. As such, chl a from these other organisms could increase the value hence the cause of higher biomass Also, chl a:C ration of phytoplankton is highly variable depending on factors such as temperature, nutrient availability (Geider & MacIntyre, 2002), hence biomass estimates from chl a should thus be viewed with caution. Before this study, no other studies in the system have estimated phytoplankton carbon biomass by microscopic analysis of abundance and biovolume measurements According to Tirok & Scharler (2013), chl a (mg chla/m^3) was highest in LP and lowest in CC. Phytoplankton biomass (Carbon, pg/L) recorded in this study calculated from biovolume (μm^3/L) of individual taxa was also highest in LP and least in CC. Since both studies used the same samples but different methods, both methods if applied cautiously could thus be used to analyse for biomass in phytoplankton studies. With respect to the variations in chl a:C ratio, nutrients, light and temperature have been postulated to positively influence chl a:C ratio (Geider et al., 1997; Wang et al., 2008). The low chl a:C ratio in LP suggest that the phytoplankton in that region have greater cellular carbon that chlorophyll hence were not as actively photosynthesising as the phytoplankton from CC and MT. Also, due to the extreme conditions of LP, LP might contain resting stages of phytoplankton which contain little or no chl a. Biomass (carbon) will thus be under estimated in the case of LP if chl a values are used as a proxy for biomass.

4.4 Autotrophy and Heterotrophy

Variability in the balance between autotrophy and heterotrophy has important implications for the energy cycling within food webs (Bukaveckas et al., 2002). In terms of abundance, autotrophs dominated throughout the system. It should be noted that heterotrophs in this study were within the nano- and microplankton range (2–200 μm) which basically included heterotrophic dinoflagellates and ciliates. An increase in heterotrophic biomass in LP from October 2010 through September 2011 was due to the decrease in salinity making the environment more suitable for grazers (heterotrophs). Similarly, zooplankton biomass also peaked with an increase in freshwater supply with the onset of the rains in January 2011(Carrasco & Perissinotto, 2012). Generally, the system recorded a higher percentage of autotroph than heterotroph biomass within the nano- and microplankton due to the shallow nature of the system. Shallow parts of estuaries are said to have higher ratios of autotrophs to heterotrophs because there is sufficient light for photosynthesis (Kemp et al., 1997; Caffrey et al., 1998) especially if nutrients are not limiting as was the case with the system during this study. However, based on this explanation, it could be expected that LP, the shallowest part of the system to have the highest autotrophic: heterotrophic biomass ratio throughout the study which was not the case. This was also because the main heterotroph in LP was *Fabrea* which had a mean cell volume of $11.7x10^4$-$69.9x10^4$ μm^3 hence very high carbon content while the main autotroph, *Cyanothece* has a cell volume of 48.9-278.3 μm^3 hence smaller carbon content.

The months when Fabrea was present thus had a high heterotrophic carbon biomass and hence a low autotroph:heterotroph biomass ratio. Lister's point also had the highest abundance and biomass of the heterotrophic dinoflagellates *Oxyrrhis marina* and *Protoperidinium*. These dinoflagellates were present in lower amounts in MT and *Protoperidinium* was below detection limit in CC. This would also explain why heterotrophs mostly dominated in LP and not other parts of the system.

The low heterotrophic biomass in MT and CC might be a result of the presence of copepods that fed preferably on ciliates and heterotrophic dinoflagellates. The widely occurring copepod *Acartia* which has been recorded in the system before as a dominant zooplankton taxa (Grindley, 1981; Carrasco & Perissinotto, 2010) has been shown to feed preferably on ciliates and heterotrophic dinoflagellates rather than phytoplankton(Yang et al., 2009; Yang et al., 2010). Studies show that more than 70% of the total carbon ration ingested by these copepods are from ciliates and heterotrophic dinoflagellates except during diatoms blooms (which was not recorded in this study) when about 60% the total carbon ration was from phytoplankton(Yang et al., 2009; Yang et al., 2010). It is therefore possible that the heterotrophs (ciliates and heterotrophic dinoflagellates) investigated in this study were selectively being grazed upon, hence the low heterotrophic biomass.

Ciliates contributed more to the heterotrophic biomass (HB) of the system than heterotrophic dinoflagellates (HDF). The lower HDF biovolume compared to ciliate biovolume was responsible for dominance of ciliate biomass. The availability of food (phytoplankton) for the ciliates was another reason for this dominance, however, food (bacteria) availability for the proliferation of HDF could not be quantified as no data for bacteria was collected. Studies have shown that in lentic and marine systems where there are no allochthonous sources of carbon and bacteria production depends on algal derived carbon, bacteria production is 30% of phytoplankton production (Robarts & Wicks, 1990; Le, 1994; Ochs, 1995). Heterotrophic dinoflagellates thus had lesser food compared to ciliates. Also studies have shown that dinoflagellates (both autotrophic and heterotrophic) flourish in stable stratified waters (Margalef, 1978; Felip & Catalan, 2000). The fact that the system was very turbid (unstable) and not stratified could possibly be responsible for the low biomass of HDF.

The HB in this study ranges from $3.04 \times 10^7 - 4.47 \times 10^9$ pg/L and is higher than that of the Gyeonggi Bay in Korea ($8.1 \times 10^6 - 8.59 \times 10^7$ pg/L) (Yang et al., 2010).The Gyeonggi Bay is eutrophic in nature with high primary productivity and a large nutrient input, but its heterotrophic biomass which is also made up of heterotrophic dinoflagellates and ciliates is less than recorded in this study. In this respect, the St. Lucia estuarine system could possibly be more productive than other eutrophic systems. However, apart from

the Gyeonggi Bay study, no other studies were heterotrophic biomass was measured from HDF and ciliates was found to better substantiate the productivity of St. Lucia estuarine lake system in this regard. The dominance of autotrophs in the system implies there is a net source of recycled carbon in the system (Bukaveckas et al., 2002). This however, would be true only if the system has limited or no allochthonous carbon sources such that heterotrophs depend only on autochthonous carbon. Although in estuaries most nutrients come from surrounding environments through river inflow, the St. Lucia estuarine lake system at the time of sampling had very limiting riverine input and was isolated from the sea hence no major external sources of carbon. Therefore, LP had the least net organic carbon in the system while CC has the most net organic carbon.

4.5 Ciliate community structure

Apart from Froneman & McQuaid (1997) who investigated ciliates and their ecological role in the Kariega estuary in Eastern Cape Province, South Africa, no studies have been conducted on ciliates in South African estuaries. Nineteen ciliate taxa (see table 5) were found during this study of which the 15 taxa identified were cosmopolitan.

The genus *Euplotes* was the main ciliate group recorded as it had the highest species richness and abundance. This was possibly because the taxon is able to flourish in hypersaline environments and there was sufficient food especially as it not only feeds on microalgae, other protist and ciliates, but it can survive on bacteria only. Also, some species of the *Euplotes* were listed in MT and CC implying they can survive on a wide variety of salinities. However, their highest abundance was recorded in LP meaning hypersaline environments are an ambient condition for their survival. The presence of the saline tolerant ciliate *Fabrea* was enhanced by the presence of *Dunaliella* known to be its principle food source (Pandey et al., 1990). *Fabrea* has been recorded to prey specifically on *Dunaliella* in the wild and flourish in estuaries that harbour the green algae species (Post et al., 1983). However, in this system, stable isotope analysis by Carrasco & Perissinotto (2012) showed that the cyanobacterium (*Cyanothece*) was the main food source for the ciliate *Fabrea*. The high abundance of *Cyanothece* in the system which co-occurred with *Dunaliella* might have masked the importance of *Dunaliella* as the main feeding preference of *Fabrea*. The occurrence of these food sources (*Dunaliella* and *Cyanothece*) only in LP would also best explain why Fabrea was found in the northern region of the system agreeing with studies which have proven that food availability controls the presence of ciliates in aquatic system (Beaver and Crisman, 1989).

The spirotrichs (*Euplotes*, *Codonella*, *Laboea*, *Strobilidium*, *Strombidinopsis* and *Tintinnopsis*) listed in this study are typically substrate orientated hence benthic in nature (Lynn, 2010). They could also be

found associated with leaf litter in mangrove ecosystems and have been noted to represent over 30% of species diversity in sediment samples (Lynn, 2010). Although water samples were collected for this study, the ciliates present were probably benthic species resuspended into the water column representing over 52.6% of the ciliate diversity. According to Lynn (2010), Tintiniids typically comprise < 25% of the total ciliate abundance. Low densities of Tintiniids (<100 Cells/L, <18.2%) have also been recorded in the Kariega estuary of South Africa (Froneman & Mcquaid, 1997) and Scheldt estuary of Belgium (1800-2900 Cells/L, 11.3 %) (Muylaert et al., 2000). This was however not true in the Mouth during this study as Codonella and *Tintinnopsis* in MT had an abundance of 25-327 Cells/L (11%) and 37-750 Cells/L (28.3%) respectively hence 39.3% of the ciliate population. In CC however, Tintiniids had an abundance of 150-2850 Cells/L comprising 25.4% of the ciliate abundance.

The ciliate biomass was lowest in CC, followed by the mouth and highest in LP. Although food (phytoplankton) was available in MT and CC, the highest biomass for ciliates was neither in MT nor CC. This was possibly due to the presence of more ciliate predators such as the ciliates; *Didinium* and *Laboea* in CC and MT. The ciliate abundance and biomass (carbon) in this system ranged from $1.10x10^{3}$–$1.08x10^{5}$ Cells/L and $2.30x10^{6}$-$3.52x10^{8}$ pg/L respectively. These abundance and biomass values are higher than those recorded in the eutrophic highly turbid, shallow Bahia estuary of Argentina $2.0x10^{2}$–$5.2x10^{3}$ Cells/L, $5.5x10^{5}$-$8.39x10^{7}$ pg/L, tropical freshwater lakes e.g. Lake Malawi of Tanzania ($2.0x10^{2}$-$1.18x10^{4}$ Cells/L, $3.0x10^{4}$–$7.82x10^{6}$ pg/L) (Yasindi & Taylor, 2003) and lakes in the temperate regions: Lake Huron and Michigan ($2.0x10^{3}$-$1.4x10^{4}$ Cells/L, $9.9x10^{6}$ – $8.73x10^{7}$ pg/L)(Carrick & Fahnenstiel, 1990), lake Ontario ($6.86x10^{6}$ pg/L)(Taylor & Heynen, 1987). Lake Malawi was oligotrophic, showed low nutrient concentrations and low phytoplankton growth rate, hence less available food for ciliates, reasons why the abundance and biomass recorded were less than those of this study. However, biomass of ciliates in the eutrophic freshwater system, Lake Victoria of Tanzania ($2.42x10^{7}$ pg/L) (Yasindi & Taylor, 2003) was not more but within the range of those recorded in St. Lucia. This might be due to the eutrophic state of Lake Victoria, which could support higher phytoplankton growth which is food for the ciliates. The St. Lucia estuarine system is thus highly productive and can be compared to the productivity of eutrophic systems (both fresh and saline) in terms of ciliate abundance and biomass.

CONCLUSION

The St. Lucia estuarine lake system was characterized by extreme temperatures, high turbidity, low water depth and high salinities with extreme hypersalinity in the northern regions. Diatoms, dinoflagellates, green algae, cyanobacteria and ciliates were identified within the nano- and microplankton in the St. Lucia estuarine system. There was a very high spatio-temporal variation in the community structure, abundance, biovolume and biomass of these groups in the system. Many factors having relative strengths have been shown to influence the community structure of phytoplankton. However, these factors and their influences vary with the system and time, therefore a combination of local factors which are not fully understood controls the community structure. Although factors such as nutrients and temperature might have influenced the community structure in this system, the prevailing factor was salinity. The most abundant taxa (taxa contributing ≥10% of the total abundance per date) per site changed with time and although species richness was high per site, only few taxa (≤5) at a time accounted for ≥10% of the nano- and microplankton abundance per each site at each date.

The changes in nano- and microplankton species occurrence have been shown to influence their biomass (carbon) as different groups vary in biovolume and carbon content. The species composition also influences the community structure of the primary heterotrophs in the system which has a direct and or indirect influence on the community structure and survival of other species up the food chain. The northern embayments had the highest heterotrophic biomass compared to South Lake and the estuarine mouth hence the northern embayment had the least net organic carbon available in the system. Overall, the St. Lucia estuarine system at the time of sampling was a very eutrophic system from the amount of nano- and microplankton biomass. Also, the estuarine mouth and South Lake had more available food for grazers as their main groups (diatoms and green algae) are a more preferred food source for phytoplankton grazers compared to cyanobacteria which was the main group in the northern embayments.

Although the microscopy method of determining abundance and biomass was very tedious and time consuming compared to a chl a method of determining algae biomass, the former method allows for more in depth reference to the species or taxa and the biomass they account for with respect to time and space. This is advantageous as the chl a method only gives values with respect to size classes if samples are size-fractionated and not to the actual taxa. It is recommended that samples for microscopic analysis be analysed soon after field collection to ease identification.

Fifteen new diatom genera never recorded by previous studies in the system were recorded in this study. Also, the eight green algae (chlorophytes), one cryptophyte, seven cyanobacteria and six dinoflagellates listed in this study have not been recorded by previous studies in the system. The differences in physico-

chemical conditions and the state of the estuarine mouth best explains the difference in the nano- and microplankton community structure recorded overall in the St. Lucia estuarine lake system.

Recommendations for future studies

Phytoplankton community structure (taxonomic composition, abundance, biovolume and biomass) changes with changing physico-chemical conditions of a system. Monthly samples analysed in this study might have missed major changes in the community structure within the month. Analysis of the samples collected within the monthly samples would give a more comprehensive understanding of the dynamics of the nano- and microplankton of the system.

An increase in precipitation in 2013 has decreased salinities and all parts of the system have approximately the same salinity values ranging from about 6-10. During this study, all sites had significantly different salinities and significant differences in nano- and microplankton composition, abundance, biovolume and biomass. An understanding of the dynamics of the nano- and microplankton of the system in its present (2013) state of lower and uniform salinities would lead to the understanding of the system both in hypersalinity and low salinity.

Also, seasonal differences in phytoplankton community structure could not be conclusive based on this study as seasonal replicates were not available. Sampling for more than a year should be conducted to better establish a trend between seasons.

APPENDIX

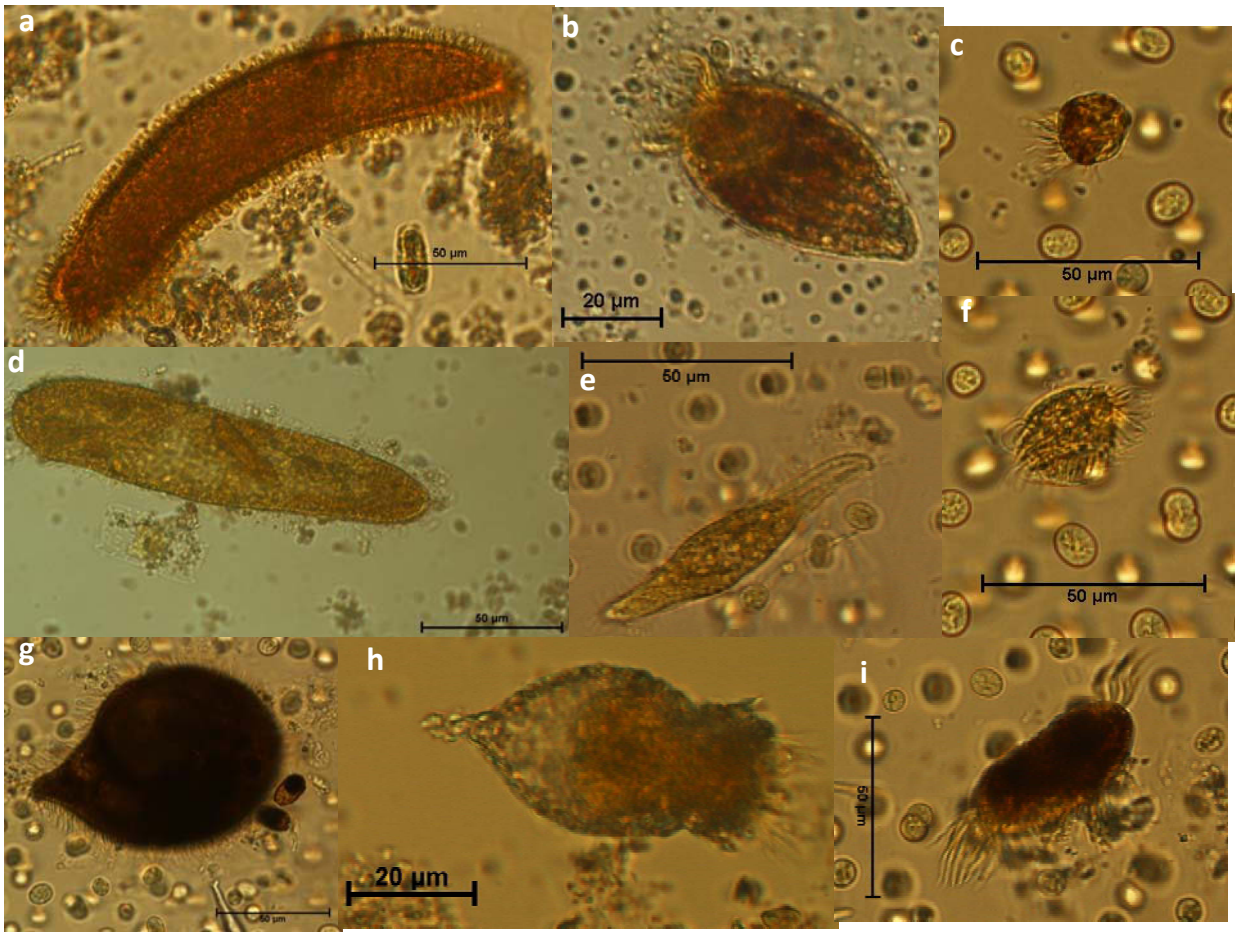

Plate 1: Some ciliates recorded during this study. a= Unknown 4, b= *Strombidinopsis* sp., c= *Euplotes* sp2, d= *Nassula* sp. e= Unknown 1, f= *Euplotes* sp4, g= *Fabrea* sp., h= *Codonella* sp. and i= *Euplotes* sp3.

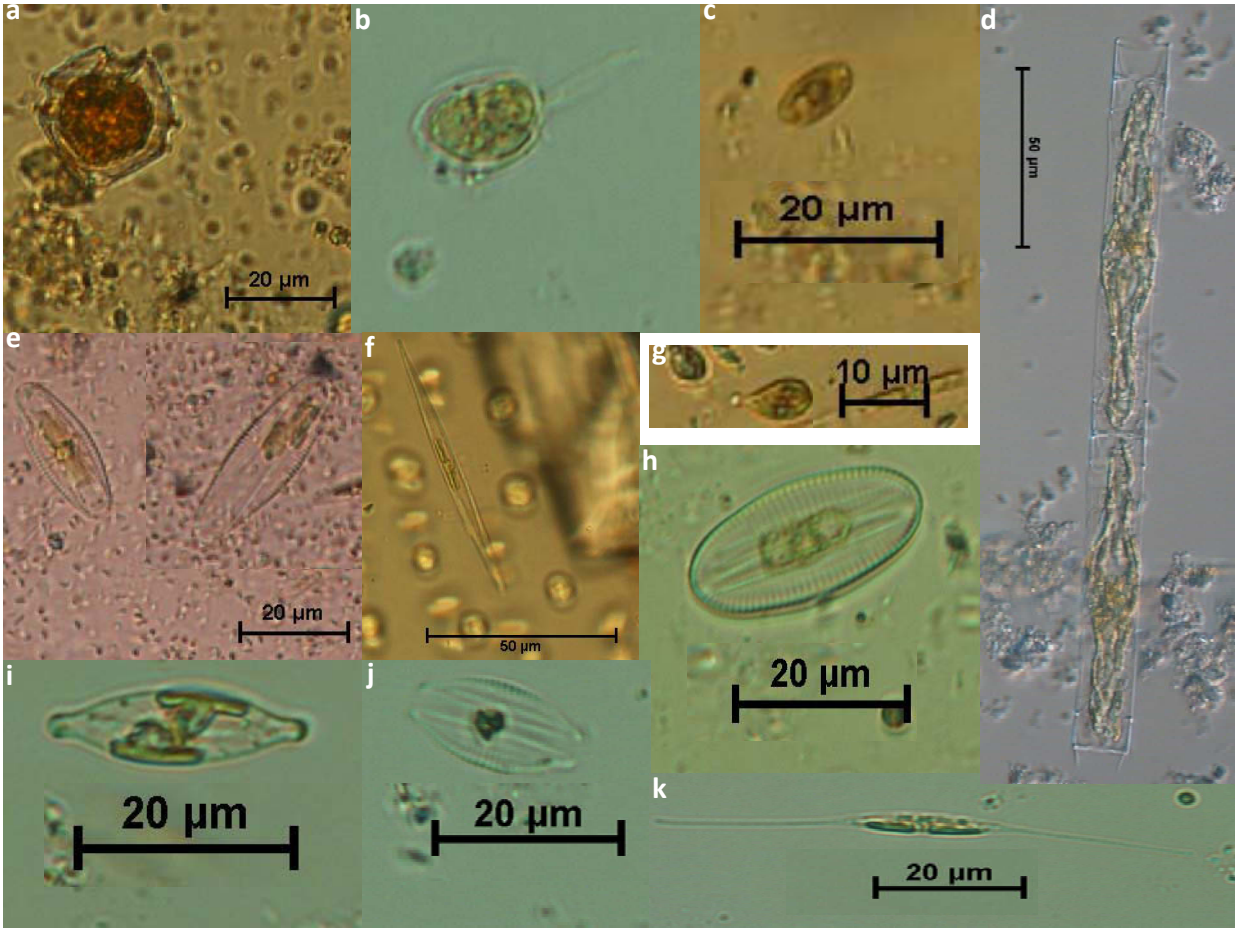

Plate 2: Some of the dominant phytoplankton taxa recorded during this study. a= *Protoperidinium* sp., b= *Prorocentrum* sp., c= *Chlamydomonas* sp., d= *Cerataulina* sp., e= *Seminavis* sp., f= *Nitzschia* sp1, g= *Chroomonas* sp., h= *Diploneis* sp2, i= *Navicula* sp2, j= *Amphora* sp. and K= *Nitzschia longissima*.

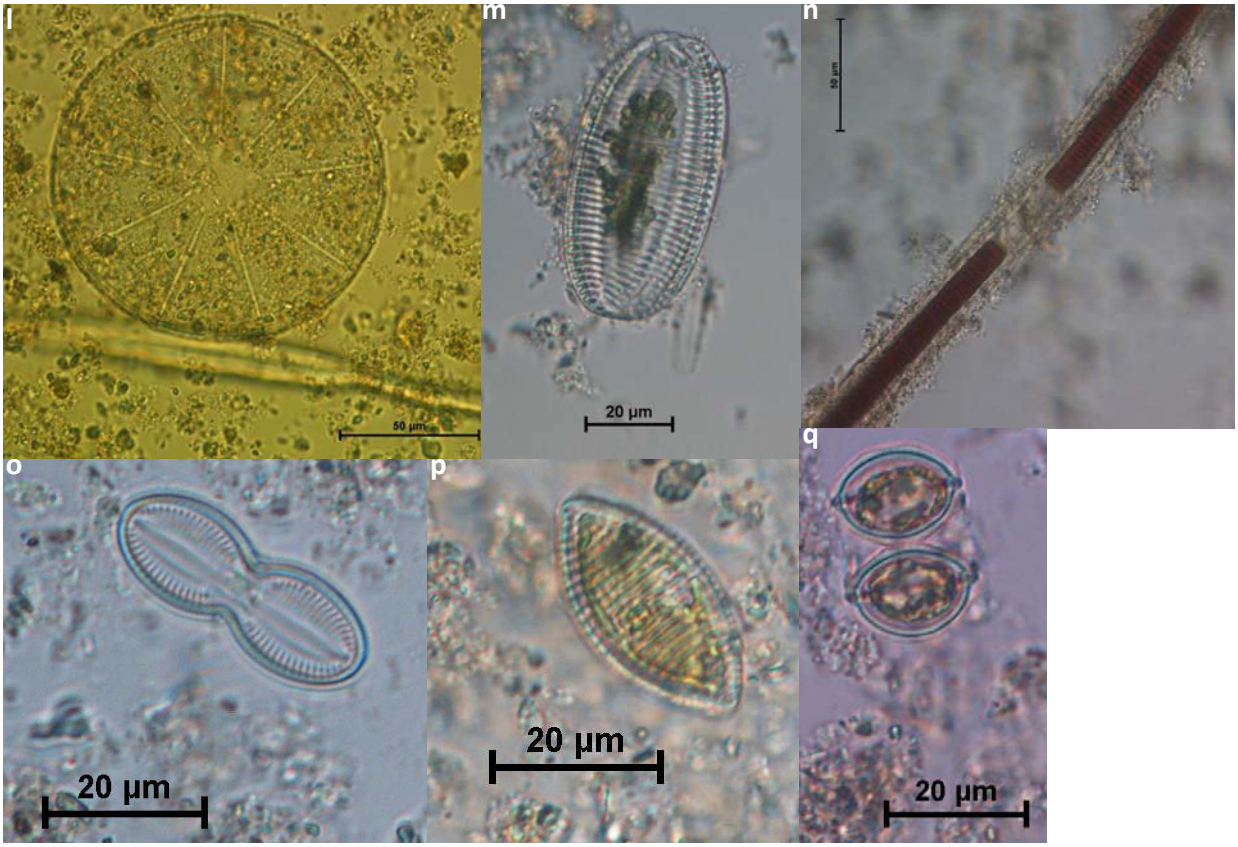

Plate 3 Continuation of some abundant taxa recorded during this study. l= *Actinoptychus* sp., m= *Tryblionella* sp., n= *Lygnbya* sp., o= *Diploneis* sp1, p= *Nitzschia* sp u2 and q= *Melosira* sp.

REFERENCES

Adams, J., Attwood, C., Bate,. Belcher, T., Berliner, D., et al., 2011. National biodiversity assessment 2011 National Estuary Biodiversity Plan for South Africa Technical Report, Cape Town.

Adams, J. and Bate, G., 1999. Primary Producers. Estuarine microalgae. In Allanson, B.R. and Baird, D. ed. *Estuaries of South Africa*. UK: Cambridge University Press, pp. 91–100.

Adolf, J.E., Yeager, C.L., Miller, D.W., Mallonee, M.E. and Harding, L.W., 2006. Environmental forcing of phytoplankton floral composition, biomass, and primary productivity in Chesapeake Bay, USA. *Estuarine, Coastal and Shelf Science*, 67, pp.108–122.

Allanson, B.R. and Baird, D., 1999. *Estuaries of South Africa*, UK: Cambridge University Press.

Allanson, B.R. and winter, D., 1999. Chemistry. In Allanson, B.R and Baird, D. ed. Estuaries of South Africa. UK: Cambridge University Press, pp. 356.

Al-rasheid, K.A.S. and Sleigh, M.A., 1995. Distribution and Abundance of Interstitial Ciliates in Southampton Water in Relation to Physicochemical Conditions , Metal Pollution and the Availability of Food. *Estuarine, Coastal and Shelf Science*, 41,pp.61–80.

Anandraj, A., Perissinotto, R. and Nozais, C., 2007. A comparative study of microalgal production in a marine versus a river-dominated temporarily open/closed estuary. South Africa. *Estuarine, Coastal and Shelf Science*, 73, pp.768–780.

Anderson, M.J., Gorley, R.N. and Clarke, K.R., 2008. PERMANOVA+ for PRIMER: Guide to software and statistical methods. ed 1. UK: PRIMER-E Limited, pp. 214.

Azémar, E.P., Boulétreau, S., Lionard, M., Muylaert, K., Vyverman, W., Meire, P. and Tracking, M., 2007. Looking for general trends in trophic interactions among estuarine micro- and mesozooplankton. *Journal of Plankton Research*, 29, pp.1135–1147.

Bate, G. and Smailes, P., 2008. The response of the diatom flora of St Lucia Lake and estuary, South Africa, to a severe drought. *African Journal of Aquatic Science*, 33), pp.1–15.

Beaver, J.R. and Crisman, T.L., 1989. The role of ciliated protozoa in pelagic freshwater ecosystems. *Microbial Ecology*, 17, pp.111–136.

Begg, G.M., 1978. Estuaries of Natal. In *Natal Town and Regional Planning Commission*. Pietermaritzburg.

Borja, A. and Basset, A., 2012. Classifying ecological quality and integrity of estuaries. In Wolanski, E. and McLusky, D. eds, Treatise on estuarine and coastal sciences. Wathman Academic Press. pp.125-162.

Bukaveckas, P.A., Williams, J.J. and Hendricks, S.P., 2002. Factors regulating autotrophy and heterotrophy in the main channel and an embayment of a large river impoundment.*Aquatic Ecology,* 36, pp.355–369.

Caffrey, J.M., Cloern, J.E. and Grenz, C., 1998. Changes in production and respiration during a spring phytoplankton bloom in San Francisco Bay, California, USA: implications for net ecosystem metabolism. *Marine Ecology Progress Series*, 172. pp.1–12.

Carrasco, N.K. and Perissinotto, R., 2012. Development of a halotolerant community in the St. Lucia Estuary (South Africa) during a hypersaline phase. *PloS one*, 7, p.e29927.

Carrasco, N.K. and Perissinotto, R., 2010. The comparative diet of the dominant zooplankton species in the St Lucia Estuary, South Africa. *Journal of Plankton Research*, 33, pp.479–490.

Carrick, H.J. and Fahnenstiel, G., 1990. Planktonic protozoa in lakes Huron and Michigan: seasonal abundance and composition of ciliates and dinoflagellates. *Journal of Great Lakes Research*, 16. pp.319–329.

Case, M., Enide, E., Leitao, S.N. and Sant Anna, E.E. Schwamborn, R. and de Moraes Junior, A.T., 2008. Plankton community as an indicator of water quality in tropical shrimp culture ponds. *Marine Pollution Bulletin*, 56, pp.1343–1352.

Chakraborty, P., Acharyya, T. and Babu, P.V.R., 2011. Impact of salinity and pH on phytoplankton community in a tropical freshwater system : An investigation with pigment analysis by HPLC. , 13, pp.1–17.

Cholnoky, B.J., 1968. Die Diatomeenassoziationen der Santa-Lucia-Lagune in Natal (Sudafrika). *Botanica Marina*, 11, pp.1–127.

Cyrus, D.P., Vivier, L. and Jerling, H.L., 2010. Effect of hypersaline and low lake conditions on ecological functioning of St Lucia estuarine system, South Africa: An overview 2002–2008. *Estuarine, Coastal and Shelf Science*, 86, pp.535–542.

Cyrus, D., Jerling, H., MacKay, F. and Vivier, L., 2011. Lake St Lucia, Africa's largest estuarine lake in crisis: Combined effects of mouth closure, low levels and hypersalinity. *South African Journal of Science*, 107, pp.1–13.

Day, J.H., 1981. *Estuarine Ecology*, Cape Town: A A Balkema.

Day, J.H., Millard, N.A.H. and Broekhuysen, G.H., 1954. The ecology of South African estuaries. Part V. The St. Lucia system. *Transaction of Royal Society of South Africa*, 34, pp.129–157.

Day, J.W., Crump, B.C., Kemp, W.M and Yàñez-Arancibia, A. 2013. Estuarine Ecology. ed. 2. New Jersey. John Wiley & Sons Publishing, pp. 537.

Dijkman, N.A. and Kromkamp, J.C., 2006. Photosynthetic characteristics of the phytoplankton in the Scheldt estuary: community and single-cell fluorescence measurements. *European Journal of Phycology*, 41, pp.425–434.

Du, Y., Xu, ., Warren, A., Lei, Y. and Dai, R., 2012. Benthic ciliate and meiofaunal communities in two contrasting habitats of an intertidal estuarine wetland. *Journal of Sea Research*, 70, pp.50–63.

Felip, M. and Catalan, J., 2000. The relationship between phytoplankton biovolume and chlorophyll in a deep oligotrophic lake : decoupling in their spatial and temporal maxima, 22, pp.91–105.

Fielding, P.J., Forbes, A.T. and Demetriades, N.T., 1991. Chlorophyll concerntrations and suspended particulate loads in St Lucia, a turbid estuary on the coast of South Africa. *South African Journal Of Marine Science*, 11, pp.491–498.

Finkel, Z.V., Beardall, J., Flynn, K.J., Quigg, A., Rees, T.A.V. and Reven, J.A., 2010. Phytoplankton in a changing world: cell size and elemental stoichiometery. *Journal of Plankton Research*, 32, pp.119–137.

Flameling, I.A. and Kromkamp, J., 1994. Responses of respiration and photosynthesis of Scenedesmus protuberans Fritsch to gradual and steep salinity increases. *Journal of Plankton Research*, 16(12), pp.1781–1791.

Forbes, A.T. and Cyrus, D.P., 1993. Biological effects of salinity gradient reversals in a southeast african estuarine lake. *Netherlands Journal of Aquatic Ecology*, 27, pp.483–488.

Forbes, A.T. and Demetriades, N.T., 2005. A review of the commercial , shallow water penaeid prawn resource of south africa. Ezemvelo KZN Wildlife, pp. 64.˜

Froneman, P.W., 2002. Responds of plankton to 3 different hydrological phases of the Kasouga Estuary. South Africa. *Estuarine, Coastal and Shelf Science*, 55, pp.535–546.

Froneman, P.W. & McQuaid, C.D., 1997. Preliminary Investigation of the Ecological Role of Microzooplankton in the Kariega Estuary , South Africa. pp.689–695.

Geider, R.J, MacIntyre, H.L. and Kana, T.M., 1997. Dynamic model of phytoplankton growth and acclimation : responses of the balanced growth rate and the chlorophyll a : carbon ratio to light , nutrient-limitation and temperature. *Marine Ecology Progress Series*, 148, pp.187–200.

Geider, R.J. and MacIntyre, H.L., 2002. Physiology and Biochemistry of Photosynthesis and Algal Carbon Acquisition. In Williams, P.J., Thomas, D.N and Reynolds, C.S. ed. Phytoplankton Productivity. Carbon assimilation in marine and fresh water ecosystems. Oxford. UK: Blackwell Publishing, pp. 44–70.

Gosselain, V., Hamilton, P.B. and Descy, J., 2000. Estimating phytoplankton carbon from microscopic counts: an application for riverine systems. *Hydrobiologia*, 438, pp.75–90.

Grange, N and Allanson, B.R., 1995. The influence of freshwater inflow on the nature, amount and distribution of seston in estuaries of the Eastern Cape. South Africa. *Estuarine, Coastal and Shelf Science*, 40, pp.403–420.

Grindley, J.R., 1981. Estuarine plankton. In Estuarine Ecology with particular reference to Southern Africa. pp. 117–146.

Grindley, J.R. and Heydorn, A.E.F., 1970. Red water and associated phenomena in St. Lucia. *South African Journal of Science*, 66, pp.210–213.

Grinienė, E., Mažeikaitė, S. and Gasūinaitė, Z.R., 2011. Inventory of the taxonomical composition of the plankton ciliates in the Curonian Lagoon (SE Baltic Sea). *International Journal of Oceanography and hydrobiolgy*, 40, pp.86–95.

Hallegraeff, G.M., Bolch, C.J.S., Hill, D.R.A., Jameson, I., LeRio, J.-M., McMinn, A., Murray, S., de Salas, M.F. and Saunders, K., 2010. Algae of Australia. Phytoplankton of Temperate Coastal Waters., Melbourne: CSIRO Publishing.

Haris, G.P., 1986. Phytoplankton Ecology. Structure, Function, and Fluctuations, London: Chapman and Hall Ltd.

Hasle, G.R., 1978. The inverted-microscope method. In Phytoplankton manual. UNESCO Monogr. Oceanogr. Method, pp. 89–96.

Van Heerden, I.., 1976. The geology of Lake St. Lucia and some aspects of its sedimentation. In A.E.F. Heydorn, ed. St. Lucia Scientific Advisory Council. Proceedings of a meeting held at Charters Creek 15-17 Febuary, 1976.

Helsinki Commission., 2006. Biovolumes and Size-Classes of Phytoplankton in the Baltic Sea.Baltic Sea Environment Proceedings No.106.

Hill, J.B., 1975. The Origin of Southern African Coastal Lakes. *Transactions of Royal society of South Africa*, 41, pp.225–239.

Hillebrand, Helmut, and Kirschtel, D., 1999. Biovolume calculation for pelagic and benthic microalgae. *Journal of Phycology*, 35, pp.403–424.

Hilmer, T., 1990. Factors influencing the estimation of primary production in small estuaries. PhD Thesis., University of Port Elizabeth.

Hilmer, T. and Bate, G.C., 1991. Vertical migration of a flagellate-dominated bloom in a shallow South African estuary. *Botanica Marina*, 34, pp.113–121.

Hocky, P. and Turpie, J., 1999. Estuarine birds of South Africa. In D. Allanson, B.R. and Baird, ed. Estuaries of South Africa. pp. 235–268.

Hoppenrath M., E.M. and D.G., 2009. Marine Phytoplankton, Germany: Kliene Senckenberg-Reihe 49.

Huang, L., Jian, W.J., Song, X.Y., Huang, X.P., Liu, S., Qian, P.Y, Yin, K.D and Wu, M. 2004. Species diversity and distribution for phytoplankton of the Pearl River estuary during rainy and dry seasons.*Marine Pollution Bulletin*,49, pp.588–596.

Hutchison, I.P.G., 1976. The hydrology of the St. Lucia system. In Heydorn A.E, ed. St. Lucia Scientific Advisory Council. Proceedings of a meeting held at Charters Creek 15-17 Febuary, 1976. Oceanographic Research Institute.

Johnson, I.M., 1977. A study of the phytoplankton of the St. Lucia system. MSc thesis, University of Natal.

Kemp, W.M., Smith, E.M., Marvin-DiPasquale, M. and Boynton, W.R., 1997. Organic carbon balance and net ecosystem metabolism in Chesapeake Bay. *Marine Ecology Progress Series*, 150, pp.229–248.

Kibirige, I. and Perissinotto, R., 2003. In situ feeding rates and grazing impact of zooplankton in a South African temporarily open estuary. *Marine Biology*, 142, pp.357–367.

Kibirige, I., Perissinotto, R. and Nozais, C., 2002. Short communication. Alternative food sources of zooplankton in a temporarily-open estuary : evidence from 13 C and 15 N.*Journal of Plankton Research*, 24, pp.1089-1095

Konoplya, B.I. and Soares, F.S., 2011. New Geometric Models for Calculation of Microalgal Biovolume. *Brazilian Archives of Biology and Technology*, 54, pp.527–534.

Kotsedi, D., 2011. The response of microalgal biomass and community composition to environmental. *Nelson Mandela Metropilitan University*, p.158.

Kotsedi, D., Adams, J.B. and Snow, G.C., 2012. The response of microalgal biomass and community composition to environmental factors in the Sundays Estuary. *Water SA*, 38, pp.177–190.

Kriel, J.P., 1966. Report of the commission of enqury into the alleged threat to plant and animal life in the St. Lucia Lake, Pretoria: Government Printer.

Krompkamp, J and Underwood, G.J.C., 1991. Primary production by phytoplankton and microphytobenthos in estuaries. *Advance in Ecological Research*, 29, pp.93–153.

Largier, J.L. and Taljaard, S., 1991. The dynamics of tidal intrusion, retention, and removal of seawater in a bar-built estuary. *Estuarine, Coastal and Shelf Science*, 33, pp.325–338.

Le, J., Wehr, J.D. and Campbell, L., 1994. Uncoupling of bacterioplankton and phytoplankton production in fresh waters is affected by inorganic nutrient limitation. *Applied and environmental microbiology*, 60, pp.2086–2093.

Lecce-monteroni, P., 2007. Biovolume determination of phytoplankton guilds in transitional water ecosystems of Mediterranean Ecoregion. *Transitional Waters Bulletin*, 2, pp.83–102.

Lee, J.J., Crockett, L.J., Hagen, J. and Stone, R.J. 1974. The taxonomic identity and physiological ecology of *Chlamydomonas hedleyi* sp. nov. algal flagellate symbiont from the foraminifera *Archaias angulatus*. *British Phycological Journal*, 9, pp 407-422.

Lemoalle, J.F. and Talling, J., 1998. Ecological dynamics of tropical waters, Cambridge: Cambridge University Press.

Lynn, D.H., 2010. The Ciliated Protozoa. characterisation, classification and guide to literature. 3rd Edition, New York: springer Science.

Margalef, R., 1978. Life-forms of phytoplankton as survival alternatives in an unstable environment. *Oceanologica Acta*, 1, pp.493–509.

Margheri, Maria C., Bosco, M., Giovannetti, L. and Ventura, S., 1999. Assessment of the genetic diversity of halotolerant coccoid cyanobacteria using amplified 16S rDNA restriction analysis. *FEMS Microbiology Letters*, 173, pp.9–16.

Marra, J., 2002. Approaches to the measurements of plankton production. In Williams D.T., Thomas D.N and Reynolds C.S., ed. Phytoplankton Productivity. Carbon assimilation in marine and fresh water ecosystems. Oxford. UK: Blackwell Publishing, pp. 78–105.

McGill, G.A., 1980. Evaluating the effectiveness of the Mfolozi-St. Lucia link by numerical modelling. Report No. 3/80., Johannesburg: Hydrological Research Unit. Univeristy of Witswatersrand.

Menden-Deuer, S., Lessard, E.J. and Satterberg, J., 2001. Effect of preservation on dinoflagellate and diatom cell volume and consequences for carbon biomass predictions. *Limnology and Oceanography*, 222, pp.41–50.

Menden-Deuer, S. and Lessard, E.J., 2000. Carbon to volume relationship for dinoflagellas, diatoms and other protist plankton. *Limnology and Oceanography*, 45, pp.569–579.

Millard, N.A.H. and Broekhuysen, G.J., 1970. The ecology of South Africa estuaries. Part X: St. Lucia: a second report. *Zoologica Africana*, 5, pp.277–307.

Mitra, A., Banerjee, K. and Gangopadhyay, A., 2004. Introduction to marine plankton, Delhi: Daya Publishing House.

Van der Molen, J.S. and Perissinotto, R., 2011. Microalgal productivity in an estuarine lake during a drought cycle: The St. Lucia Estuary, South Africa. *Estuarine, Coastal and Shelf Science*, 92, pp.1–9.

Montagnes, David, J.S., Berges, John, A., Harrison, Paul, J., and Taylor, F.J.R., 1994. Estimating carbon , nitrogen, protein and chlorophyll a from volume in marine phytoplankton. *Limnology and oceanography*, 39, pp.1044–1060.

Moss, B., Booker, I., Balls, H. and Monson, K., 1989. Phytoplankton distribution in a temperate floodplain lake and river system I. Hydrology, nutreint sources and phytoplankton biomass. *Journal of Plankton Research*, 11, pp.813–838.

Muir, D.G. & Perissinotto, R., 2011. Persistent phytoplankton bloom in Lake St. Lucia (iSimangaliso Wetland Park, South Africa) caused by a cyanobacterium closely associated with the genus Cyanothece (Synechococcaceae, Chroococcales). *Applied and environmental microbiology*, 77, pp.5888–96.

Muylaert, K., Riet Van, M., Koen, S., Tackx, M., and Vyverman, W., 2000. Dynamics and trophic roles of heterotrophic protists in the plankton of a freshwater tidal estuary. *Hydrobiologia*, 432, pp.25–36.

National Research Council, 1969. Recommended procedures for measuring the productivity of plankton standing stock and related oceanic properties, Washington D.C: National Academy of Science.

O'Boyle, S. and Silke, J., 2010. A review of phytoplankton ecology in estuarine and coastal waters around Ireland. *Journal of Plankton Research*, 32, pp.99–118.

Ochs, C.A., Cole, J.J., and Likens, G.E., 1995. Population dynamics of bacterioplankton in an oligotrophic lake. *Journal of Plankton Research*, 17, pp.365– 391.

Oksanen, J.F., Blanchet, G., Kindt, R., Legendre P., Minchin, P.R., O'Hara, R. B., Simpson, G.L., Solymos, P.M., Stevens, H.H. and Wagner, H., 2013.vegan: Community Ecology Package. R package version 2.0-9.http://CRAN.R-project.org/package=vegan

Oren, A., 2005. A hundred years of Dunaliella research: 1905-2005. *Saline systems*, 1, pp.1-14.

Orme, R.A., 1973. Barrier and lagoon systems along the Zululand coast, South Africa. In Coastal Geomorphology. Coates, R.D. ed., Binghampton, New York: State University of New York.

Pandey, B.D., Yeragi, S.G. and Pal, A.K., 1990. Nutritional Value of a Heterotrichous Ciliate, Fabrea salina with Emphasis on Its Fatty Acid Profile.*Asian Australian Journal of Animal Sciences*, 17, pp.995–999.

Paxinos, R. and Mitchell, J.G., 2000. A rapid Utermöhl method for estimating algal numbers. *Journal of Plankton Research*, 22, pp.2255–2262.

Perissinotto, R., Bate, G.C. and Muir, D.G., 2013. Microalgae. In R. Perissinotto, R., Stretch, D.D. and Taylor, ed. Ecology and Conservation of Estuarine Ecosystems. Lake St Lucia as a Global Model. Cambridge: Cambridge University Press, pp. 198–207.

Perissinotto, R., Pillay, D. and Bate, G., 2010. Microalgal biomass in the St Lucia Estuary during the 2004 to 2007 drought period. *Marine Ecology Progress Series*, 405, pp.147–161.

Perissinotto, R., Nozais, C., Kibirige, I. and Anandraj, A., 2003. Planktonic food webs and benthic-pelagic coupling in three South African temporarily-open estuaries. *Acta Oecologica*, 24, pp.307–316.

Pfister, G., Auer, B. and Arndt, H., 2002. Pelagic ciliates (Protozoa, Ciliophora) of different brackish and freshwater lakes -a community analysis at the species level. *Limnologica*, 32, pp.147–168.

De Phillipis, R., Margheri, M.C., Pelosi, E. and Ventura, S., 1993. Exopolysaccharide production by a unicellular cyanobacterium isolated from a hypersaline habitat. *Journal of Applied Phycology*, 5, pp.387–394.

Pillay, D. and Perissinotto, R., 2009. Community structure of epibenthic meiofauna in the St. Lucia Estuarine Lake (South Africa) during a drought phase. *Estuarine, Coastal and Shelf Science*, 81, pp.94–104.

Post, F.J., Borowitzka, L.J., Borowitzka, M.A., Mackay, B. and Moulton, T., 1983. The protozoans of a Western Australian hypersaline lagoon. *Hydrobiologia*, 105, pp.95–113.

Potter, I.C., Chuwen, B.M., Hoeksema, S.D. and Elliott, M., 2010. The concept of an estuary□: A definition that incorporates systems which can become closed to the ocean and hypersaline. *Estuarine, Coastal and Shelf Science*, 87, pp.497–500.

Putt, M. and Stoecker, D.K., 1989. An experimentally determined carbon: volume ratio for marine oligotrichous ciliates from estuarine and coastal waters. *Limnology and Oceanography*, 34, pp.1097–1103.

R Core Team. 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.URL http://www.R-project.org/.

Ramdani, M., Elkhiati, N., Flower, R.J., Thompson, J.R., Chouba, L., Kraiem, M.M., Ayache, F. and Ahmed, M.H., 2009. Environmental influences on the qualitative and quantitative composition of phytoplankton and zooplankton in North African coastal lagoons. *Hydrobiologia*, 622, pp.113–131.

Redden, Anna, M. and Rukminasari, N., 2008. Effects of increases in salinity on phytoplankton in the Broadwater of the Myall Lakes, NSW, Australia. *Hydrobiologia*, 608, pp.87–97.

Redfield A.C., 1934. On the proportions of organic derivatives in sea water and their relation to the composition of plankton. R. J. Daniel, ed., Liverpool: University Press, Liverpool.

Reynolds, C.S., 2006. Ecology of Phytoplankton, UK: Cambridge University Press.

Rippka Rosmarie, Deruelles Josette, Waterbury John, B., Herdman Micheal and Stanier Roger, Y., 1979. Generic Assignments, Strain Histories and Properties of Pure Cultures of Cyanobacteria. *Journal of General Microbiology*, 111, pp.1–61.

Robarts, R.D. and Wicks, R.J., 1990. Heterotrophic bacterial production and its dependence on autotrophic production in a hypertrophic African reservoir. *Journal of Fisheries and Aquatic Sciences*, 47, pp.1027–1037.

Round, F.E., Crawford, R.M. and Mann, D.G., 1990. The diatoms, Great Britian: Cambrige University Press.

Roussomoustakaki, M. and Anagnostidis, K., 1991. Cyanothece halobia, a new planktic chroococcacean cyanophyte from Hellenic heliothermal salt works. *Archiv für Hydrobiologie, Supplement, Algological studies*, 64, pp.71–95.

Schumann, E., Lagier, J. and Slinger, J., 1999. Estuarine hydrodynamics. In B. Allanson and D. Baird, eds. Estuaries of South Africa. UK: Cambridge University Press, pp. 27–52.

Sherr, E.B. and Sherr, B.F., 1994. Bacterivory and herbivory: Key roles to phagotrophic protist in pelagic food webs. *Microbial Ecology*, 28, pp.223–235.

Snow, G.C., 2000. Structure and Dynamics of Estuarine Microalgae in the Gamtoos Estuary. MSc dissertation. *University of Port Elizabeth.*, pp 104.

Šorf, M. and Brandl, Z., 2012. The rotifer contribution to the diet of Eudiaptomus gracilis (G. O. Sars, 1863) (Copepoda, Calanoida). *Crustaceana*, 85, pp.1421–1429.

Struski, C. and Bacher, C. 2006. Preliminary estimate of primary production by phytoplankton in Marennes-Oleron Bay, France. *Estuarine, Coastal and Shelf Science*, 66, pp.323-334.

Sun, J., 2003. Geometric models for calculating cell biovolume and surface area for phytoplankton. *Journal of Plankton Research*, 25, pp.1331–1346.

Suthers, I.M. and Rissik, D. 2009 Plankton: A guide to their ecology and monitoring for water quality. Australia: Csiro Publishing, pp. 256

Taylor, R.H., 2006. Ecological responses to changes in the physical environment of the St Lucia Estuary. PhD Thesis, Department of Plant and Environmental Sciences, Norwegian University of Life Sciences,.

Taylor, R.H., 1991. The Greater St. Lucia Wetland Park. Natal Parks Board.

Taylor, W.D. and Heynen, M.L., 1987. Seasonal and vertical distribution of ciliophora in lake Ontorio. *Aquatic Ecosystem Health and Management*, 6, pp.289–297.

Thompson, P.A., Guo, M. and Harrison, P.J., 1992. Effects of variation in temperature. 1. On the biochemical composition of 8 species of marine phytoplankton. *Journal of Phycology*, 28, pp.481–488.

Tillmann, U., Heesse, K., Colijn, F., 2000. Planktonic primary production in the German Wadden Sea. Journal of Plankton Research 22, 1253-1276.

Tirok, K. and Scharler, U.M., 2013. Dynamics of pelagic and benthic microalgae during drought conditions in a shallow estuarine lake (Lake St . Lucia). *Estuarine, Coastal and Shelf Science*.

Tomas, R.C., 1997. *Identifying Marine Phytoplankton*, New York: Academic Press.

Tyrell, T., 1999. The relative influences of nitrogen and phophorus on oceanic primary production. *Nature*, 400, pp.525–531.

Urrutxurtu, I., Orive, E. and de la Sota, A., 2003. Seasonal dynamics of ciliated protozoa and their potential food in an eutrophic estuary (Bay of Biscay). *Estuarine, Coastal and Shelf Science*, 57, pp.1169–1182.

Wang, X. J., Behrenfeld, M., Le Borgne, R., Murtugudde, R. and Boss, E., 2008. Regulation of phytoplankton carbon to chlorophyll ratio by light, nutrients and temperature in the equatorial Pacific Ocean: a basin-scale model. *Biogeosciences Discussions*, 5(5), pp.3869–3903.

Wasserman, R.J., Noyon, M., Avery, T.S. and Froneman, W.P., 2013. Trophic level stability-inducing effects of predaceous early juvenile fish in an estuarine mesocosm study. *PloS one*, 8(4), p.e61019.

Welschmeyer N.A., 1994. Fluorometric analysis of chlorophyll chlorophyll b and pheopigments. *Limnology and oceanography*, 39, pp.1985–1992.

Welsh, Eric A., Liberton, M., Stöckel, J., Loh, T., Elvitigal, T., Wang, C., Wollam, A., Fulton, R.S., Clifton, S.W., Jacobs, J.M., Aurora, R., Ghosh, B.K., Sherman, L.A., Smith, R.D., Wilson, R.K and Pakrasi, H.B., 2008. The genome of Cyanothece 51142 , a unicellular diazotrophic cyanobacterium important in the marine nitrogen cycle. *Proceedings of the National Acedamy of Sciences, U S A.*, 105, pp.15094–9.

Whitfield, A.K., 1977. Predation on fish in lake St. Lucia, Zululand. MSc Thesis, Pietermaritzburg: University of Natal.

Whitfield, A.K., 1992. The characterisation of South African Estuarine Systems, Pretoria: Water Research Commision.

Whitfield, A.K. and Blaber, S.J.B., 1978. Feeding ecology of piscivorous birds at the Lake St. Lucia. Part II: Wadding Birds, Part III: Swimming Birds. *Ostrich*, 50, pp.10–20.

Whitfield, A.K. and Taylor, R.H., 2009. Case studies and reviews A review of the importance of freshwater inflow to the future conservation of Lake St Lucia. *Aquatic Conservation: Marine and Freshwater Ecosystems*.19, pp.838-848.

Wimpenny, R.S., 1966. *The plankton of the sea*, Russell Square London: Faber and Faber Ltd.

Yang, E.J., Ju, S.-J. and Choi, J.-K., 2010. Feeding activity of the copepod Acartia hongi on phytoplankton and micro-zooplankton in Gyeonggi Bay, Yellow Sea. *Estuarine, Coastal and Shelf Science*, 88, pp.292–301.

Yang, E.J., Kang, H.-K., Yoo, S. and Hyun, J.-H., 2009. Contribution of auto- and heterotrophic protozoa to the diet of copepods in the Ulleung Basin, East Sea/Japan Sea. *Journal of Plankton Research*, 31, pp.647–659.

Yasindi, A.W. and Taylor, W.D., 2003. Abundance , Biomass and Estimated Production of Planktonic Ciliates in Lakes Victoria and Malawi. *Aquatic Ecosystem Health & Management*, 6, pp.289–297.