On the effects of wastewater effluent on local primary production near a Canadian Arctic coastal community

by

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ABSTRACT

The Arctic Ocean is experiencing large and small changes throughout the region caused by climate-induced change. Furthermore, increases in human activity and population have been observed throughout the Arctic, leading to an increase in wastewater production within coastal Arctic communities. Wastewater contains high concentrations of nitrogen compounds that are released during summer into coastal seas, via natural purification systems such as lagoons and wetlands, where it can stimulate marine algal production. My thesis research investigated the effect of this anthropogenic nitrogen input on phytoplankton near an Arctic coastal community in the Canadian Arctic Archipelago, namely Cambridge Bay, Nunavut. During discharge, a phytoplankton bloom was triggered by the presence of high nitrogen compounds in the effluent, which influenced both taxonomic composition and production of the phytoplankton community. Before wastewater discharge, flagellate species were dominant in the phytoplankton community, characteristic of the original oligotrophic conditions for the region. However, after discharge commenced, diatom rapidly became the dominant taxa of the bloom. The increase in diatoms was also believed to influence bloom progression as the diatoms were dense enough to sink below a persistent pycnocline and use accumulated nutrients available at depth in the system. A comparison of observations before and after wastewater discharge versus those during shows that the average primary production increased by about 170 mg C m^{-2} d^{-1} during discharge. This increase over a 4-week period equated to 68% of total open water primary production from 9 July to 10 October, 2018. Although the local system was not deemed eutrophic, a persistent pycnocline and bounding shallow sills for the local bay create a unique situation that acts to trap wastewater nitrogen compounds at depth. Such a system can lead to increasing stores of nutrients over time if not regularly ventilated. Therefore, strong recommendations are made for future monitoring and research.

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I would like to express my sincere gratitude to my supervisor, Professor C.J. Mundy. He has provided me with a lot of research opportunities and has always given me the support and advice I needed. Thanks to his continued interest, affection, and teaching, the days of my study were truly enjoyable and a lot of progress has been made. One of the things I am the most grateful for in my life is that I had been granted an opportunity to have him as my supervisor.

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DEDICATION

My grandmother always used to tell me: “You are my pride”

Even though I have never told you this, your dedication to taking care of me and the fact that I was making you proud were the seeds from which everything I have accomplished grew.

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CHAPTER ONE: INTRODUCTION

1.1 Introduction

Concerns of the public all around the world associated with ongoing global warming are rising. Of particular concern is the Arctic, where temperatures are rising at more than twice the global rate (AMAP 2017). The Arctic Ocean is strongly affected by anthropogenic stressors, including indirect impacts associated with climate-induced change and direct impacts through local human activities (Häder and Gao, 2015). In general, the Arctic Ocean has a lower annual primary production than that of more temperate regions due to nitrogen depletion as well as light limitation associated with polar darkness, snow and sea ice cover, and strong surface stratification (Harrison and Platt, 1986; Harrison and Cota, 1991). Climate-induced change is causing sea ice to start melting earlier, to a greater extent, and to commence freezing later (Stroeve et al., 2014). The major impacts of this change are increases in open-water periods, ocean temperature, and light transmission into the water column, which has triggered an overall increase in net primary production as determined by satellite observations (Bélanger et al., 2013; Arrigo and Van Dijken, 2015).

Additionally, the declining ice-covered period in the Arctic, caused by the ongoing global warming, has made the area more accessible for tourism, transportation, and natural resource extraction, which have led to an increase in human population in the Arctic territories (Crépin et al., 2017), especially Canada's northern territory. Accordingly, it is expected that an increasing population will influence the production and discharge of wastewater from coastal Arctic communities and into the adjacent marine system (Chouinard et al., 2014). Resultant anthropogenic nutrients and eutrophication effects can stimulate phytoplankton and their production in coastal zones (Falkowski, 1994; Yang et al., 2008).

Studies on changes in the physical, chemical and biological conditions of the Arctic Ocean and the construction of forecasting systems have been actively conducted. Many related studies (e.g., Bélanger et al., 2013; Stroeve et al., 2014; Arrigo and Van Dijken, 2015) have primarily focused on the increase in net primary production caused by the increase of light availability in the water column due to the decrease of the amount of sea ice. However, the fact that the nutrients affect phytoplankton growth as much as light, should not be overlooked (Harrison and Cota, 1991). To determine the trend of changes in the Arctic Ocean and net primary production caused by ongoing global warming, changes in nutrient concentrations, ratios and dynamics should be carefully analyzed while keeping in mind light availability in the Arctic Ocean. It can be inferred that net primary production in coastal Arctic oceans is affected by an increased presence of wastewater effluent caused by increases in the industrial presence and an increase in the human population in the region. Of particular concern is the potential for anthropogenic eutrophication and the potential for harmful algal blooms, which can produce toxins that can have deleterious effects on local ecosystems with implications on human health (Scholin et al., 2000). However, due to a lack of available information about the impact of anthropogenic stressors on Arctic regions, there is an urgent need to better understand the influence of wastewater outflow on nutrient enrichment near coastal Arctic communities and their impact on local primary production. Although many studies have effectively examined the impact of wastewater effluent on estuaries in temperate regions (e.g., Pan and Rao, 1997; Yang et al., 2008), no distinctive findings related to wastewater effluent in Arctic coastal regions yet have been published.

Much of the Canadian Arctic is considered oligotrophic, where primary production has the strong potential to be enhanced by nutrient addition to surface waters, and particularly

nitrogen (Ardyna et al., 2011; Tremblay et al., 2015). With the current development trends in the Arctic and associated climate-induced changes in marine ecosystems, the influx of anthropogenic nitrogen compounds into the Arctic coastal region could have a critical impact on the ecosystem through enhancement of phytoplankton production (i.e., eutrophication). In this thesis, I examine the impact of wastewater effluent on phytoplankton species composition and local primary production through a comparison of seasonal versus anthropogenic variability in phytoplankton biomass, community composition and production in Cambridge Bay. In the next section, I outline my specific thesis objectives.

1.2 Thesis Objectives

The overarching goal of my thesis research is to assess the impact of wastewater outflow on local phytoplankton community composition and production in the coastal Arctic regions through a case study in Cambridge Bay, Nunavut, Canada. Cambridge Bay is located within a shallow and relatively fresh waterway of the western Northwest Passage called the Kitikmeot Sea. A secondary goal to place observations into a regional context is to examine interannual variability of key physical and environmental variables in the Kitikmeot Sea. More specifically, my research objectives are to:

1. Quantify the interannual and spatial variability of temperature, salinity, and nutrient concentrations in the Kitikmeot Sea.
2. Describe the processes driving seasonal progression of stratification, nutrient concentration and phytoplankton biomass in Cambridge Bay; and
3. Assess the impact of wastewater associated with nutrient enrichment on phytoplankton biomass, community composition and production.

1.3 Thesis Structure

My thesis is put together as a sandwich-style thesis with a single main research chapter to be submitted as a manuscript for peer-review in an international journal. This manuscript encompasses results for all objectives. My thesis chapters include 1: Introduction, 2: Background, 3: Manuscript, and 4: Conclusion and Recommendations. In Chapter 1 of this thesis, the high-level importance of the thesis research is provided as well as the thesis objectives and the structure. In Chapter 2, background information on the coastal marine ecosystem and wastewater treatment systems used in Arctic regions is described. Particular emphasis of this description is placed on the lagoon-wetland treatment system and environmental characteristics in Cambridge Bay, Nunavut, Canada, the location of my thesis research. Chapter 3 presents a manuscript with the following citation,

Back, D.Y., Ha, S., Else, B., Hanson, M., Jones, S., Shin, K., Tatarek, A., Wiktor, J.M., Cicek, N., Alam, S., and Mundy, C.J. (*In* preparation). The impact of wastewater effluent on phytoplankton in the Arctic coastal zone: A case study in the Kitimeot Sea of the Canadian Arctic. To be submitted to *Environment International*.

The manuscript encapsulates a 6-year spatial dataset in the Kitikmeot Sea from 2013 to 2018, while providing a detailed biological oceanography case study of the 2018 phytoplankton production period within Cambridge Bay. I note that I am the primary author of the manuscript and led all aspects of the manuscript and associated research, with the assistance of my co-authors and particularly, my thesis advisor, Dr. C.J. Mundy. Finally, the thesis is concluded in Chapter 4 and accompanied by suggestions for future research in Cambridge Bay and other coastal Arctic communities.

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CHAPTER TWO: BACKGROUND

2.1 Limiting and Influencing Factors on Phytoplankton Growth

Limiting and influencing factors of phytoplankton growth in the Arctic Ocean include light and nutrient availability, temperature and salinity, respectively (Harrison and Cota, 1991; Gosselin et al., 1997; Carmack et al., 2004; Olli et al., 2007). Light limitation arguably has the greatest control on phytoplankton production in the Arctic Ocean (Harrison and Platt, 1986; Sommer and Lengfellner 2008; Lewandows et al., 2010). This statement is supported by the overall increase in phytoplankton production across with the increasing ice-free season influenced by the warming Arctic climate (Rysgaard et al., 1999; Bélanger et al., 2013; Arrigo and Van Dijken, 2015). Outside of the polar darkness period, light limitation influences photosynthesis during the ice-covered period and at depth after the ice breaks up because of the formation of a deep chlorophyll maximum (DCM; **Figure 2.1**; Leu et al., 2015). Prevalence of a DCM throughout the Canadian Arctic has shown that phytoplankton tend to locate at depths just above the 1% level of photosynthetically active radiation (PAR), balanced by nutrient limitation in surface waters during summer (Martin et al., 2010). During summer, nitrogen is the main limiting nutrient in surface waters of the Arctic Ocean (Codispoti et al., 2013; Tremblay et al., 2015); however, the relative concentration of dissolved inorganic silica to nitrogen can influence taxonomic composition of the phytoplankton community (Egge and Aksnes, 1992). Temperature influences phytoplankton growth in terms of its metabolic rate (Arrigo et al., 2010; Lewandowska et al., 2010). Although lower temperatures generally cause lower growth rate in most organisms, many Arctic species of phytoplankton have adapted to the decreased temperature range of the Arctic Ocean (Harrison and Cota 1991; Lewandowska et al., 2010) For instance, the temperature of maximum growth for polar microalgae is between 4°C and 14°C

(Kottmeier and Sullivan, 1988; Arrigo et al., 2010). Lastly, due to ice freeze and melt cycles as well as extensive riverine input into the Arctic Ocean, phytoplankton can experience some osmotic changes throughout an annual cycle (Bonilla et al., 2009). Furthermore, the significant freshwater input from riverine and ice melt sources leads to strong stratification during summers, which significantly affects nutrient dynamics (Carmack and Wassmann, 2006).

2.2 Light Availability

The amount of light, more specifically photosynthetically active radiation (PAR; 400-700 nm), in upper ocean has a great influence on the phytoplankton population and photosynthetic rates (Sverdrup, 1953). Beer 's law can be used to explain light transmission to depth in the water column:

$$I_z = I_0 e^{-K_d z} \quad [2.1]$$

Where I_z is the irradiance (µmol m^{-2} s^{-1}) transmitted to depth z (m), I_0 is the incident irradiance just under the surface, and K_d is the downwelling attenuation coefficient (m^{-1}). In particular, K_d explains light transmission to depth as an exponential decay, and thus, PAR is exponentially more available towards the surface of the ocean. The euphotic zone can be described at the depth of 1% and even 0.2% I_0 (Falkowski, 1994), the prior observed to match the depth of the DCM in Canadian Arctic waters (Martin et al., 2010). More recently, an isolume corresponding to the depth at which daily integrated PAR is 0.415 mol m^{-2} d^{-1}, has been used to describe the depth extent of phytoplankton production (Oziel et al., 2019; Matthes et al., 2020). The isolume depth tends to match the euphotic depth in late spring and summer, but often decouples during fall in the Arctic as polar darkness sets in. Therefore, the isolume is believed a more accurate description of the euphotic depth during late fall in the Arctic. Compared to temperate regions, the Arctic ecosystem has less light availability, due to the presence of snow-covered sea ice well

into spring. Phytoplankton blooms tend to commence under the sea ice during advanced melt stages when ample PAR transmits to depth under the sea ice (Mundy et al., 2014; Oziel et al., 2019). During summer, 24 daylight leads to ample light availability for production; however, surface nutrient depletion causes the bloom to be a balance of nutrient and light limitation as it settles to light limiting depths as a DCM (Martin et al., 2010).

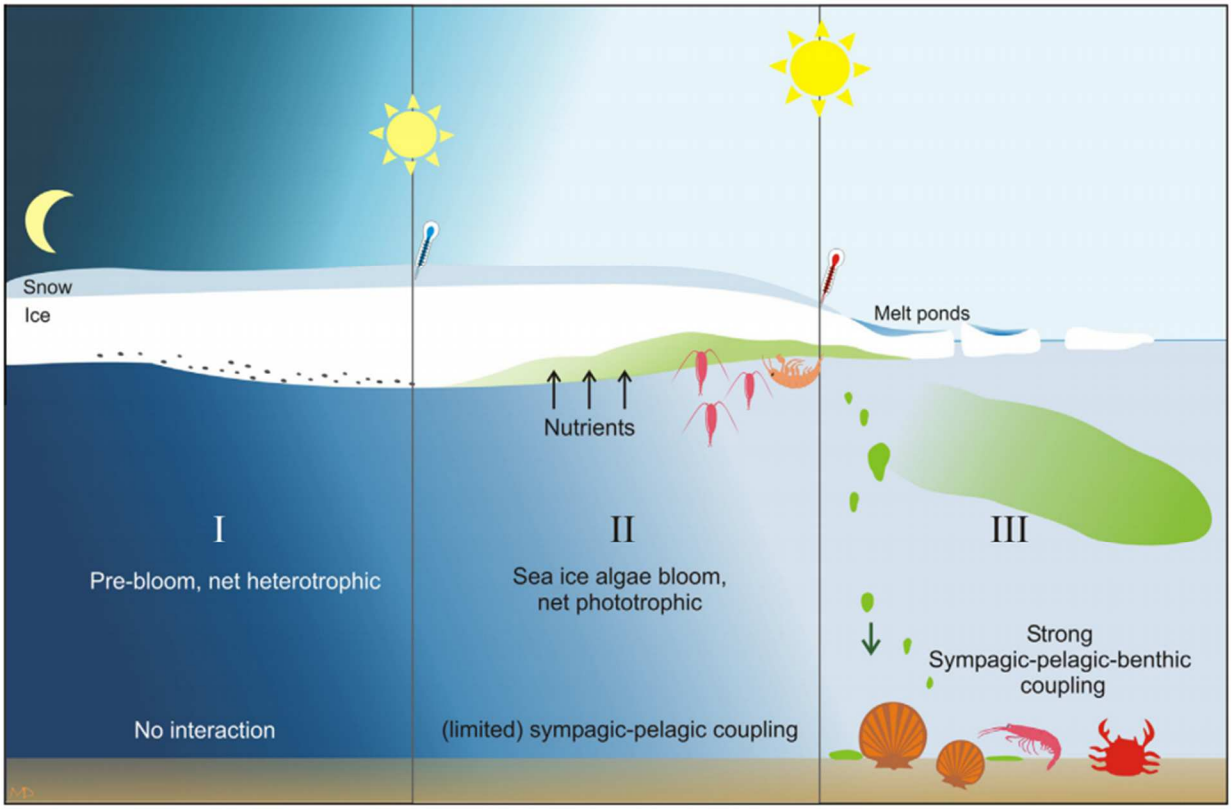

Figure 2.1 The process of ice and phytoplankton blooms during winter-summer transition divided by three major phases. The change in the first and second phase shows that there are increases in light availability, while nutrients in late winter are replete. The third phase occurs as air temperature increases, the sea ice starts melting, and a phytoplankton bloom commences under the ice. Rapidly surface nutrients are depleted by the bloom resulting in a DCM near the timing of ice break-up. From Leu et al. (2015). Copyright (2020) Elsevier.

Climate-induced changes are increasing light availability for phytoplankton across the Arctic Ocean, through decreases in ice thickness of sea ice and the period of ice cover (Stroeve et al., 2014), as well as increases in the transparency of melting sea ice (Horvat et al., 2017). The latter is due to an increase in melt pond fraction in the Arctic associated with younger and flatter ice (Kwok and Rothrock, 2009; Nicolaus et al., 2012; Rösel and Kaleschke, 2012), creating a more favourable environment for phytoplankton blooms under the ice cover. In fact, large phytoplankton blooms underneath the sea ice have been observed in the Arctic Ocean (Fortier et al., 2002; Mundy et al., 2009; Arrigo et al., 2012; Mundy et al., 2014; Oziel et al., 2019).

2.3 Nutrient Distribution and Dynamics

In the ocean, nutrient ratios vary from region to region (Geider and LaRoche, 2002), but tend to follow the Redfield stoichiometry on average. Redfield (1958) discovered that the proportion of dissolved nutrients in the deep sea is quite similar to that of nutrient composition of phytoplankton due to the role of remineralization of phytoplankton exported vertically (Arrigo, 2005). The Redfield ratio, updated to include Si for diatoms, is C:N:Si:P = 106:16:15:1 for natural phytoplankton assemblages (Brzezinski, 1985).

According to their nutrient status, oceans can be divided into eutrophic and oligotrophic waters. Ratios of available nutrients play an important role in determining the nutrient status in the waters (Geider and La Roche, 2002). Through phytoplankton culture experiments in the laboratory, Geider and La Roche (2002) were able to classify oligotrophic waters with nitrogen-limitation when the N: P ratio was less than 5. Indeed, many regions of the Canadian Arctic are considered oligotrophic and nitrogen-limited, such as the eastern Beaufort Sea and central archipelago (Ardyna et al., 2011). However, much of the Canadian Arctic, and rest of the Arctic Ocean, have molar ratios of N:P ranging from 11 to 16 (**Figure 2.2**, Tremblay et al., 2015).

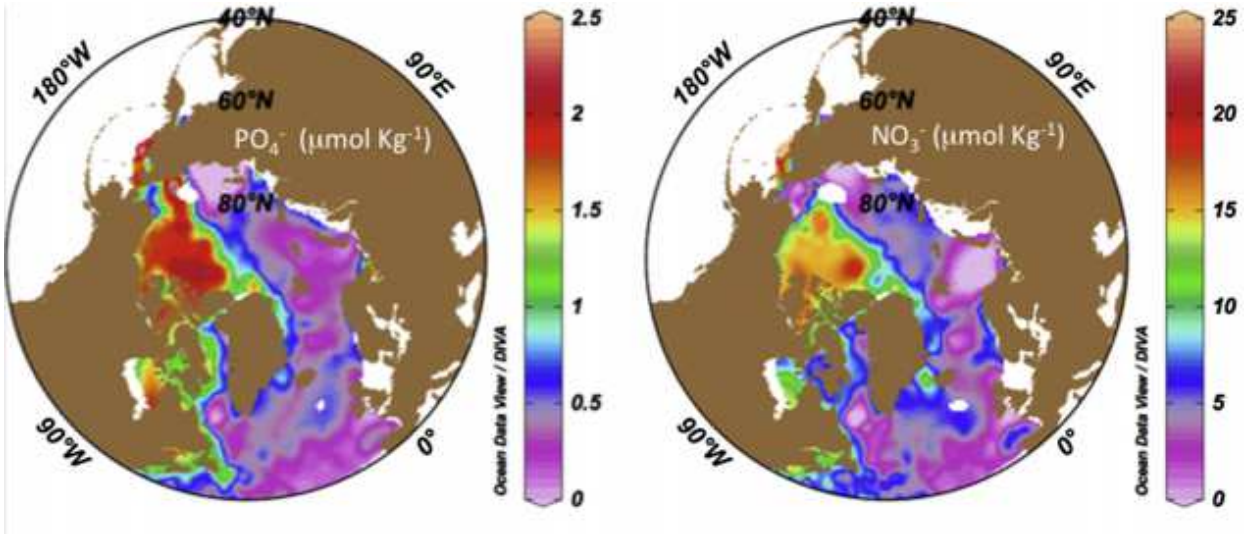

Figure 2.2. Horizontal distributions of nutrients along potential surface density in the Arctic Ocean. From Tremblay et al. (2015). Copyright (2020) Elsevier.

In contrast to phosphorus-limited freshwater systems, nitrogen concentrations and dynamics in the Arctic Ocean are closely related to primary production, since most Arctic Ocean regions have limited amounts of nitrogen in the surface layer but are relatively phosphorus-replete (**Figure 2.2**; Codispoti et al., 2013; Tremblay et al., 2015). Such nitrogen depletion in the surface layer is closely associated with strong stratification affected by low salinity caused by the input of riverine and sea ice melting (**Figure 2.3**; Rudels et al., 1991; Codispoti et al., 2013; Tremblay et al., 2015), while higher concentrations of NO_3^- are present at depths below the euphotic zone/nitracline (Sherr et al., 2003; Tremblay et al., 2015). Phytoplankton communities rapidly consume the winter-replete surface nutrients during blooms in spring, ultimately forming the DCM, which, as mentioned above, balances the requirements for light from above and nutrients from below (Carmack et al., 2004; Martin et al., 2010).

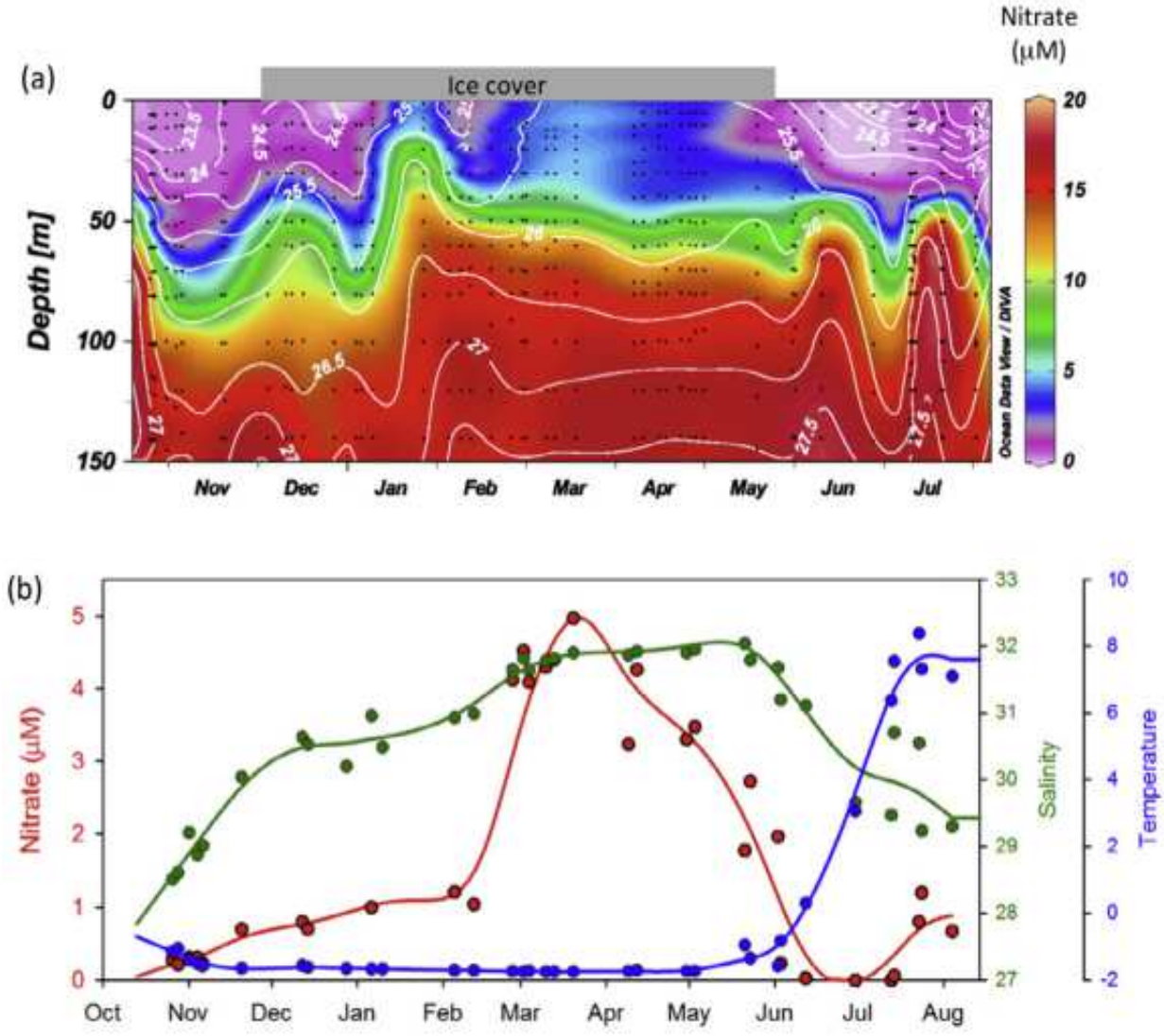

Figure 2.3. An example annual time series of (a) NO_3^- concentrations with depth and (b) surface NO_3^- concentration (red), salinity (green), and temperature (blue) in the southeast Beaufort Sea from 2007 to 2008. From Tremblay et al. (2015). Copyright (2020) Elsevier.

Inorganic nitrogen undergoes a series of physical and biologically-mediated transformations in oceanic and aquatic ecosystems. Important precesses include nitrification, assimilation and remineralization (Fouilland et al., 2007; Granger et al., 2011; Tremblay et al., 2015). The mechanisms of the nitrogen cycle in the Arctic Ocean are as follows: (1) Nitrification is a process in which ammonium (NH_4^+) is oxidized to nitrite (NO_2^-) and nitrate (NO_3^-), which

are produced by archaea and nitrifying bacteria (Tremblay et al., 2015; Fan et al., 2015). This process mainly occurs in the upper halocline and the upper mixed layer in the Arctic (Tremblay et al., 2015). (2) Algae and many microorganisms assimilate NH_4^+, NO_2^-, and NO_3^- into organic matter (OM) (Arrigo, 2005), which directly affects nitrogen availability and thus the proportion of regenerated primary production in the Arctic (Tremblay et al., 2015). (3) All organisms and ultimately microbes contribute to the process of remineralization, i.e., the breakdown and conversion of OM back into inorganic nutrients (Bourgeois et al., 2017). Indeed, remineralization in the Arctic is directly related to nutrient availability after the dark winter period (Codispoti et al., 2013).

2.4 Stratification and Mixing

Carmack and Wassmann (2006) defined different oceans based on whether temperature or salinity gradients had a greater influence on water column stratification (**Figure 2.4**). In general, oceans from temperate to tropical climates are defined as α -oceans, which are driven by temperature stratification, often with inverse salinity gradients (i.e., greater salinity towards the surface) due to evaporation. However, as temperatures decrease, salinity changes have a greater influence on density changes in the water column. Therefore, sub-Arctic and Arctic oceans tend to be salinity stratified, or β-oceans, often with inverse temperature gradients (i.e., increase in temperature with depth) due mainly to warmer yet saltier Atlantic water found at depth in the Arctic Ocean (**Figure 2.4**; Carmack and Wassmann, 2006). The β characteristic of the Arctic Ocean is further strengthened by ice melt and riverine freshwater supply (Carmack and Wassmann, 2006). The resultant strong stratification impedes mixing, which can limit primary production due to a finite nutrient supply in the surface mixed layer (**Figure 2.3**; Codispoti et al., 2013; Carmack et al., 2016). During fall, strong winds and storms frequently occur in the Arctic,

which can act to deepen and mix in new nutrients into the surface mixed layer. This process can lead to the phenomenon of fall blooms (Tremblay et al., 2002; Michel et al., 2006). Due to later freeze-up, fall blooms appear to be increasing in frequency in the Arctic Ocean (Ardyna et al., 2014). Furthermore, some parts of the Arctic Ocean have fast ocean currents and high tides, which can augment mixing and supply primary producers with nutrients (Cota et al., 1996; Dalman et al., 2019). During winter, ice formation and associated dense brine rejection can act to additionally mix surface waters, deepening and in some cases, breaking down surface stratification. This mixing also acts to increase new nutrient supply to the surface ocean and combined with the dominant remineralization process occurring during the dark polar winter, and surface nutrients become replete (**Figure 2.3**; Tremblay et al., 2015). In this case, replete refers to a variable quantity dependent on local nutrient inventories. This end of winter/early spring period is often when nitrate is at its greatest concentration, completing the annual nutrient-production cycle.

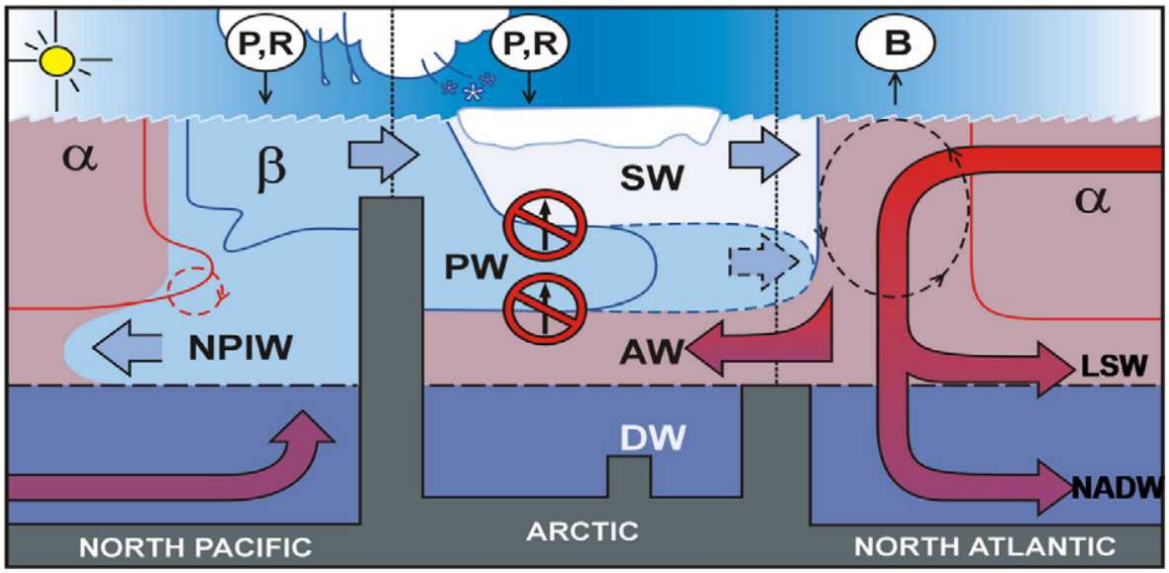

Figure 2.4. Physical oceanography related to buoyancy, stratification, and vertical mixing. Defining α and β oceans; B, buoyancy flux; P, precipitation; R, run-off. NPIW, North Pacific Intermediate Water; PW, Pacific origin water; SW, polar surface water; AW, Atlantic Water; DW, Deep Arctic Water; LSW, Labrador Sea Water; NADW, North Atlantic Deep Water. From Carmack and Wassmann (2006). Copyright (2020) Elsevier.

2.5 Phytoplankton Biomass and Community Composition

Phytoplankton community composition is closely related to primary production as well as the marine food web in the region (Lewandowska et al., 2010; Edwards et al., 2016). Therefore, it is very important to assess the phytoplankton biomass and their community composition in order to understand the marine ecosystem. Phytoplankton species composition is directly and indirectly influenced by limiting factors, i.e., light and nutrients, as well as influencing factors, i.e., temperature and salinity (Seoane et al., 2005; Yang et al., 2005; Lewandowska et al., 2010; Edwards et al., 2016). Dominant phytoplankton of a community tend to be adapted to periodic environmental changes, but their species composition can still be affected by the osmotic stress due to abrupt changes in salinity (Cota and Smith, 1991; Park et al., 2001) as well as by the

influx of anthropogenic nutrients (Yang et al., 2005).

Phytoplankton community composition has been examined using many different techniques. Microscope-based observation of phytoplankton community composition can be highly quantitative, but require significant analysis time and knowledge for accurate taxonomy, and is limited to algal cell sizes detectable by available microscope optics (often 1-5 µm in diameter; Roy et al., 2006). More recently, genomic analysis has been able to show never before realized detail in taxonomic diversity, but is limited with respect to a lack of quantitative cell abundance estimates (Vaulot et al., 2007). Another method uses pigment composition through high- performance liquid chromatography (HPLC) analysis. Although often expensive, this technique is relatively rapid and can be used to provide information on algal functional group relative abundance for even the smallest phytoplankton taxa (Seoane et al., 2005; Roy et al., 2006).

Total Chlorophyll *a*, which includes the sum of Chlorophyll *a* (Chl *a*), divinyl-Chl *a*, Chl *a* isomer and epimer, and chlorophyllide *a* (Babin et al., 2003; Bricaud et al., 2004), can be used to standardize and compare the contribution of accessory pigments used by algae to identify contribution of different phytoplankton taxa or functional groups (Jeffrey et al., 2005). However, there are different phytoplankton species that produce the same accessory pigments, and even the same phytoplankton species can differ in the relative production of each pigment from region to region (Mackey et al., 1996; Zapata et al., 2000; Jeffrey et al., 2005). Due to these limitations, it is often good to combine methods to determine phytoplankton taxonomic composition. In fact, the number of studies that use both techniques in a single study is increasing (Llewellyn et al., 2005).

Dominant species of phytoplankton and their measured quantities are frequently used as

indicators of water conditions because they are sensitive and easily changed by biotic and abiotic factors (Ardyna et al., 2011; Parmar et al., 2016). For example, eutrophic waters in the Baffin Bay, Lancaster Sound and the central Amundsen Gulf tend to be dominated by more diatom species, whereas, oligotrophic waters in the eastern Beaufort Sea, the peripheral Amundsen Gulf, and the central region of the Canadian Arctic Archipelago tend to be dominated more by flagellate species (Ardyna et al., 2011). Phytoplankton size was also influenced by the nutrient-status of waters in the Canadian Arctic Archipelago, where small picophytoplankton (0.2-2 µm) tended to dominate the most oligotrophic waters towards the western Canadian Arctic, while nano- (2-20 µm) and microphytoplankton (>20 µm) tended to dominate the more eutrophic regions towards the east (Trembley et al., 2009). Diatom species are generally bigger than flagellate species, often fitting within the microphytoplankton size range. In general, larger taxa have a greater biomass and thus require a greater nutrient supply to grow (Miller and Wheeler, 2012), which leads to the aforementioned observations based on the nutrient status of the region. If the nutrient status is changed by anthropogenic input, phytoplankton composition can change.

2.6 Primary Production

Through photosynthesis, phytoplankton absorb light with their pigments and harness the energy via their photosystems and membrane-mediated electron transport chain to produce energy for metabolism, reproduction and respiration, as well as becoming a food source for consumers (Chassot et al., 2010; Valiela, 2015). Through photosystems, adenosine diphosphate (ADP) converts to adenosine triphosphate (ATP) when the requirements of light availability are met and oxygen is produced (Lalli and Parson, 1997, Valiela, 2015). Through the Calvin-Benson cycle in the dark, produced ATP converts to ADP because this cycle makes use of phosphate and CO_2 is removed (Valiela, 2015). Furthermore, the growth and productivity of phytoplankton,

which makes up most of the primary production in marine ecosystems, depends on phytoplankton biomass and its photosynthetic rate (Falkowski, 1981; Chassot et al., 2010). In fact, phytoplankton biomass is difficult to measure directly, which has led researchers to use chlorophyll as a proxy (Falkowski, 1981; Valiela, 2015). That is, the Chl *a* pigment is used in almost all photosynthetic organisms (Seoane et al., 2005). Chl *a* is also often used to standardize carbon production measurements. For example, the carbon fixation capacity of phytoplankton can be expressed as primary production per unit of Chl *a*, which is called the assimilation number (Falkowski, 1981; Valiela, 2015). The assimilation number is used to estimate the activity of phytoplankton (Milligan et al., 2015). That is, phytoplankton are growing quickly when the assimilation number is high.

In general, the Arctic Ocean has relatively low Chl *a* concentration and biological diversity compared to lower latitude oceans (AMAP, 2009; Barton et al., 2010). Primary production in the Arctic Ocean is mainly produced by phytoplankton and ice algae, and it is controlled by the Arctic's unique physical environment and conditions (Harrison and Cota, 1991). Nutrients are supplied to primary producers through not only limited advection and convection of new nutrients (e.g., nitrate) to the surface layer in the Arctic Ocean via inflows from the Pacific or Atlantic water masses but also through regeneration of nutrients (Rudels et al., 1991; Granger et al., 2011; Codispoti et al., 2013; Tremblay et al., 2015). For instance, in **Figure 2.3b**, the increase in NO_3^- concentration from November to February is dominated by nutrient cycle such as remineralization in winter, whereas the rapid increase between February and April is more a mechanism of convection under sea ice, moving nutrients from deeper depths to surface water (*see section 2.3*). Thus, phytoplankton spring blooms in the Arctic Ocean are light-limited to start as surface nutrients are relatively replete (Leu et al., 2015). After blooms

commence, nutrient-depletion rapidly occurs in surface layers, leading to nutrient limitation, causing phytoplankton biomass and production to decrease during summer (Harrison and Cota, 1991; Carmack et al., 2016). Due to the limitation of NO_3^- concentrations in the surface layer, DCMs form closely related to nitracline, but above the depth of 1% PAR (Martin et al., 2010).

Climate-induced change is indirectly and directly affecting the Arctic marine system. Changes, such as longer open water periods and decreases in sea ice thickness, are leading to an overall increase in net primary production in the Arctic Ocean (Stroeve and Notz, 2018; Horvat et al., 2017). To exemplify, an increase in light transmission into the water column due to increase in the open-water periods has led to an increase in phytoplankton production by more than 14% per decade across most areas of the Arctic Ocean since 1998 (Bélanger et al., 2013; Arrigo and Van Dijken, 2015). Additionally, the increased open water season, yet increased freshwater input associated with increased riverine and melting sea ice can have both advantageous effects, such as increased atmosphere-ocean coupling, increased riverine nutrient supply, and increased production in the surface layers (Carmack et al., 2004; Olli et al., 2007), and disadvantageous effects, such as greater stratification, deepening nitracline, and greater nutrient limitation (Bergeron and Tremblay, 2014; Li et al., 2009), on phytoplankton communities and ecosystem. These competing effects are closely connected to the distribution of nutrients into the water column. To be more specific, extended open-water periods and mixing tend to improve net primary production at the surface until surface nutrient depletion eventually occurs (Olli et al., 2007). On the other hand, the enhanced surface stratification hinders the movement of nutrients from deeper areas to the surface (Carmack et al., 2016). Thus, earlier nutrient depletion in the surface layer can occur due to climate-induced changes (Carmack et al., 2004; Arrigo et al., 2014).

2.7 Wastewater Treatment System in the Arctic

In many Arctic regions, wastewater treatment systems are operating via natural wetlands with retention lagoons (Yates et al., 2012). These systems are based on the capacities of microbial degradation and wetland's purification, and they are affected by the local climate. The lower atmospheric and ground temperatures in the Arctic slow down the rate of microbial treatment processes, requiring longer residence times of wastewater in the lagoons and the wetland than in temperate regions (Yates et al., 2012; Chouinard et al., 2014). The result is that some wastewater is often released directly into the receiving waters (Gunnarsdóttir et al., 2013).

Arctic community wastewater collection systems are operated either through bucket toilets or flush toilets depending on the usage rates and areas (**Figure 2.5**; Gunnarsdóttir et al., 2013). The main difference between bucket and flush toilets is that the flush toilets dilute wastewater using water, so this method is characterized by lower concentrations than one that involves using a bucket (Gunnarsdóttir et al., 2013).

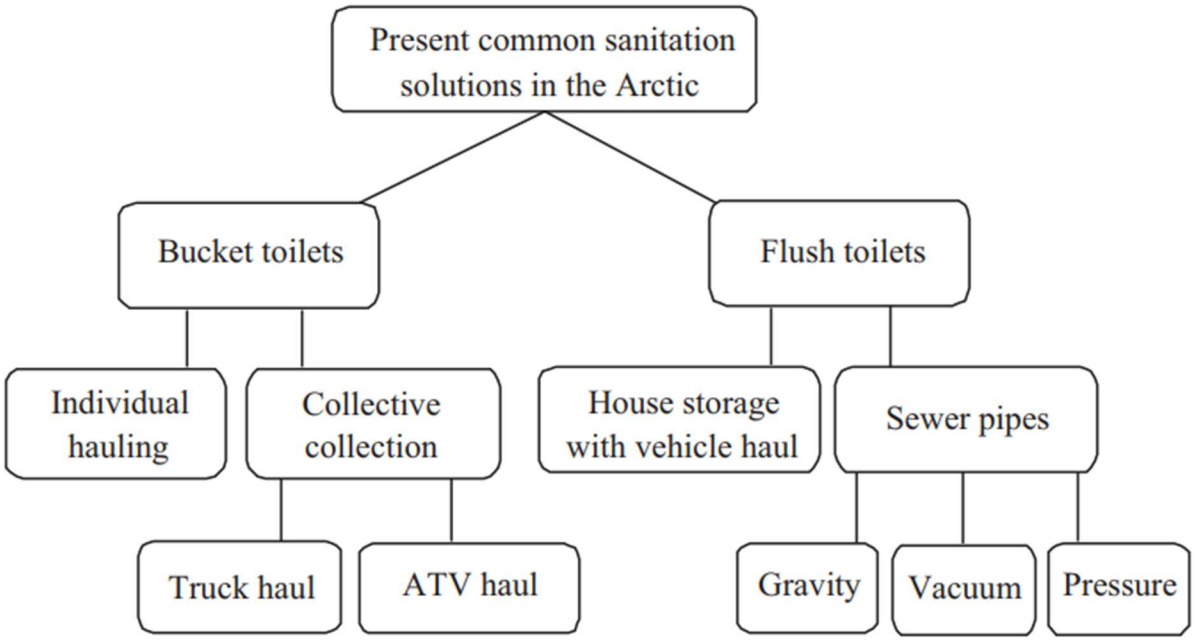

Figure 2.5. Various collection methods of wastewater from houses and camps in the Arctic. Defining ATV; All-Terrain Vehicle. From Gunnarsdóttir et al. (2013). Copyright (2020) Elsevier.

Wastewaters are almost entirely produced by human activities, and these wastewaters contain high concentrations of nitrogen compounds (Yates et al., 2012; Gunnarsdóttir et al., 2013; Chaves-Barquero et al., 2016). Therefore, as human activities increase in the Arctic region, more wastewater and related nitrogen compounds as well as some contaminants will enter the sea, affecting local chemical and biological systems in the ocean (Gunnarsdóttir et al., 2013; Chouinard et al., 2014; Chaves-Barquero et al., 2016). Such wastewater can affect the Arctic food web in a variety of ways, such as increased nutrient levels that can cause a shift in community composition and production rates, and enhance bio-accumulation and magnification of increased contaminant supply through the marine food web and acute toxicity of organisms (AMAP, 2009; Gunnarsdóttir et al., 2013).

2.8 Hydrography and Wastewater System in Cambridge Bay

Approximately 1750 people live in Cambridge Bay. The community ranks as the 6th most populated community of 25 within Nunavut in the Canadian Arctic (Government of Nunavut). The community household wastewater system uses house storage with vehicle haul in the categories of flush toilets (**Figure 2.5**), with a wastewater treatment system that operates via natural wetlands with retention lagoons. According to a recent case study of the wastewater treatment system of Cambridge Bay, almost no total nitrogen concentration changes were registered during wastewater discharge in 2014 (Chaves-Barquero et al., 2016). However, total nitrogen concentration decreased from 13.3 mg L^{-1} (wetland) to 0.4 mg L^{-1}, diluted by ocean salinities as it reached receiving waters of Cambridge Bay (Chaves-Barquero et al., 2016). 0.4 mg L^{-1} of total nitrogen concentration, which is converted to approximately 30 μmol L^{-1}, is very high compared to dissolved nutrient concentrations found in Dease Strait and also compared to almost all regions of the Arctic Ocean.

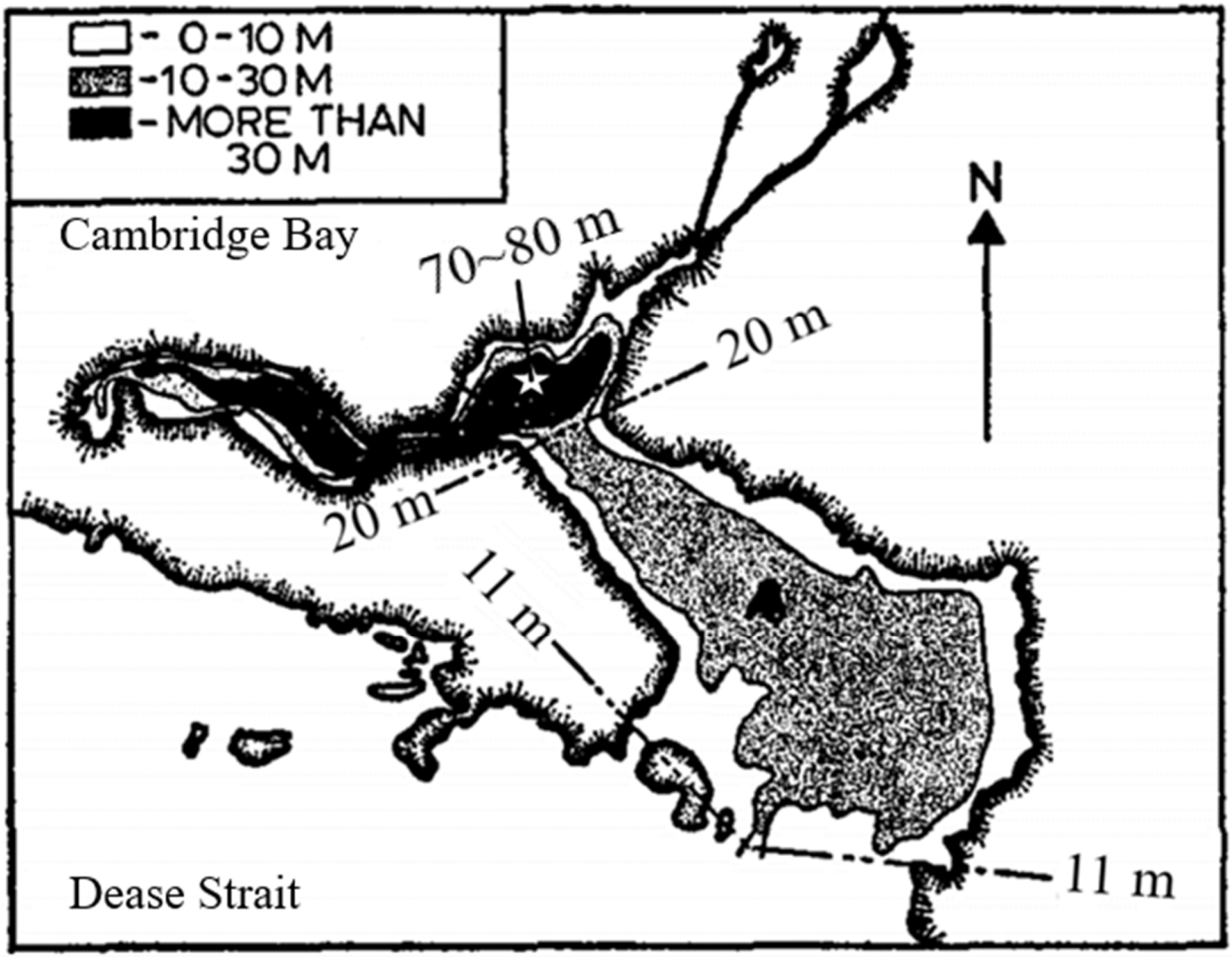

Figure 2.6. A bathymetric map of Cambridge Bay, showing locations of 20 m and 11 m sills towards the bay's exit point to the Kitikmeot Sea. The star depicts a 70-80 m deep centre of the bay. Modified from Gade et al. (1974). Copyright (2020) Elsevier.

Cambridge Bay has two very shallow sills, an outer sill of 11 m and an inner sill of 20 m, with a deep centre (**Figure 2.6**; Gade et al., 1974). This fjordal-like bathymetry can limit exchange of deepwater with the surrounding Kitikmeot Sea. This limitation in exchange is accentuated by summer runoff and ice melt that strongly stratify surface waters within Cambridge Bay (Gade et al., 1974). In turn, the bay's bathymetry and stratification will limit the exchange of inorganic nutrients via both convection (vertical) and advection (horizontal) (Carmack et al., 2016). However, some particulate organic matters (POM) is dense enough to

sink to below the stratified surface layer (Eppley and Peterson, 1979; Dorota et al., 2005; Olli et al., 2007). With the unique bathymetry of Cambridge Bay, such a mechanism could act to trap wastewater-associated nitrogen compounds at depth within the bay. As a result, the concentrations of nutrients below the surface stratification of Cambridge Bay can likely reflect the increased supply from the anthropogenic sources in the bay, limiting their mixing into the surrounding Kitikmeot Sea by environmental and bathymetric characteristics. Therefore, it can be hypothesized that many of the anthropogenic nitrogen additions will be trapped.

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CHAPTER THREE: On the impact of wastewater effluent on phytoplankton in the Arctic coastal zone: A case study in the Kitikmeot Sea of the Canadian Arctic

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Abstract

With a declining ice cover, the Arctic is also being exposed to an increasing industrial presence as it becomes more accessible for tourism, transportation, and natural resource extraction. As a result, Canada's northern territory of Nunavut is experiencing rapid population growth relative to the rest of Canada. It can be expected that an increasing population will influence the amount of wastewater production in coastal Arctic communities, that already face difficulties in wastewater system efficiencies due to the cold and dark climate. Here we present a case study on the impact of effluent from a wastewater lagoon-wetland system on local primary production near a coastal Arctic community over spring to fall 2018. Results are also placed within an interannual and regional context for the surrounding Kitikmeot Sea, located in the southwestern Canadian Arctic Archipelago. The Kitikmeot Sea is a shallow, relatively fresh sea, determined to be one of the most nutrient-deplete regions of the entire Arctic Ocean with NO_3^-+NO_2^- concentrations below the surface mixed layer rarely exceeding 2 μmol L^{-1} and a N:Si:P ratio of 1.1:7:1. The fjordal-type bathymetry of Cambridge Bay, Nunavut, and the existence of a persistent at 29 m below the bay's exit sill led to a slightly elevated N:Si:P of 2.7:10.7:1 through trapping of wastewater-sourced N at depth via sinking and remineralization of local primary production. Total production in Cambridge Bay over the 3-month open water period was 10.5 g C m^{-2} with 68% of this production occurring during the 1-month discharge of wastewater into the system. Local primary production responded rapidly to the high NO_3^-+NO_2^-, NH_4^+ and particulate organic N

(PON) concentrations provided in the wastewater effluent, accounting for up to 31% of the primary production above background rates. Remaining production was associated with the deep nutrient pool in the bay, which was only accessed towards the end of the discharge period as the diatom-dominated deep chlorophyll maximum settled below the pycnocline. Although the system is concluded to not yet be eutrophic, caution is raised at the rapid response of the marine system to the wastewater release with a strong recommendation to develop a future monitoring and research program in the bay.

3.1 Introduction

The Arctic marine ecosystem is being heavily influenced by anthropogenic stressors, including indirect impacts associated with climate change and direct through local human activities. Primarily, climate change is causing sea ice to melt earlier and to a greater extent (Stroeve et al., 2014). Major impacts of this change are increases in open-water periods and light transmission into the water column, which has triggered a >14% per decade increase in Arctic Ocean primary production since 1998, according to the satellite record (Bélanger et al., 2013; Arrigo and Van Dijken, 2015).

With climate change influencing a declining ice cover and earlier melt, the Arctic is also being exposed to an increasing industrial presence as it becomes more accessible for tourism, transportation, and natural resource extraction (Crépin et al., 2017). In part an outfall of this change, Canada's northern territory, Nunavut, is seeing a rapid population growth relative to the rest of the country (Southcott, 2014). As the human population is directly associated with the quantity and discharge of wastewater production in coastal Arctic communities, this rapid growth is leading to an increase in wastewater effluent to the aquatic environment (Chouinard et al., 2014; Gunnarsdóttir et al., 2013). Compared to more temperate regions, the harsh environment

of the Arctic influences lower effectiveness of wastewater treatment. In particular, polar darkness and prolonged freezing temperatures cause treatment mechanisms such as sorption by soil and photo-degradation to be slower and less efficient (Chouinard et al., 2014; Chaves-Barquero et al., 2016). The subsequent effluent can be relatively high in total nitrogen compounds, among other essential nutrients, that are released to the ocean where they can augment primary production in the local environment. Of particular concern is the potential for harmful algal blooms, which can produce toxins that can have deleterious effects on local ecosystems with implications on human health (Scholin et al., 2000). Due to a lack of available information about the impact of anthropogenic stressors on Arctic regions, there is an urgent need to better understand the influence of wastewater outflow on nutrient enrichment near coastal Arctic communities and their impact on local primary production.

Chaves-Barquero et al. (2016) recently investigated the chemical composition of wastewater released from the Cambridge Bay, Nunavut, wastewater lagoon treatment system. Total nitrogen (TN) concentration was only reduced from 13.5 to 13.3 mg L^{-1} within the wastewater system, which was diluted to 0.4 mg L^{-1} after being released into the local marine environment. The TN concentration of 0.4 mg L^{-1} can be converted to ~30 µmol L^{-1}, which is abnormally high relative to surface waters near Cambridge Bay that have reported early spring concentrations of 1-3 µmol L^{-1} nitrate plus nitrite (NO_3^-+NO_2^-) and ~1 µmol L^{-1} ammonium (NH_4^+) (Campbell et al. 2016; Dalman et al. 2019; Kim et al. *Submitted*). In fact, the local system, referred to as the Kitikmeot Sea, appears to be strongly nitrogen-limited, as evidenced from low production estimates (Campbell et al. 2017; Kim et al. *Submitted*), high lipid: protein allocation and low *f*-ratios (average of 0.41) for both ice algae and phytoplankton (Kim et al. *Submitted*). It is noted that the high TN concentration of the wastewater effluent reported in

Chaves-Barquero et al. (2016) can potentially be in the form of phytoplankton and microorganisms growing in the wastewater treatment system (Delgadillo-Mirquez et al., 2016). Unfortunately, dissolved inorganic nitrogen composition was not measured in the Chaves-Barquero et al. (2016) study and therefore, questions remain on the actual impact this high nitrogen content water will have on the local system.

It is hypothesized that wastewater release into the system will cause an increase in primary production within Cambridge Bay versus outside of the bay in Dease Strait. This hypothesis is made regardless of direct input of inorganic nitrogen compounds such as NO_3^- $+NO_2^-$ and NH_4^+, or indirect input via freshwater organic matter that will release a portion of the nitrogen to the inorganic pool via osmotic stress, cell death and lysis, sloppy feeding by zooplankton, and remineralization (Bronk and Steinberg, 2008). The overall aim of this paper is to investigate the impact, if any, of wastewater effluent on primary production through a case study in Cambridge Bay, Nunavut. More specifically, the study seeks to: i) quantify the interannual and spatial variability of summertime inorganic nutrient concentrations across the Kitikmeot Sea; ii) characterize seasonal phytoplankton community composition and production from spring through fall as a function of light and nutrient access; and iii) assess the impact of wastewater effluent on local primary production in the bay. Evidence from the study highlights an important localized effect of the wastewater effluent on primary production, warranting closer monitoring of the oceanic system as population numbers continue to rise.

3.2 Materials and Methods

3.2.1 Field Sites

Distributed sampling of hydrographic conditions and nutrient concentrations across the Kitikmeot Sea, Nunavut, Canada, occurred aboard a 20-m research vessel, the *R/V Martin Bergmann*, from 2013 to 2018. *S*ampling was carried out shortly after ice-break-up in the region every year. Stations were targeted for sampling covering Cambridge Bay (CB), West (WD) and East (ED) Dease Strait, and Queen Maud Gulf (QM; **Figure 3.1**). However, only in 2013 and 2018 were all sites visited in a single year due to a variety of logistics-related issues. Station sampling included hydrographic profiles followed by water sampling at the surface, 5-10 m, middle of the pycnocline (~20-40 m across the Kitikmeot Sea), chlorophyll maximum, and 1-5 m above the bottom.

In 2018, a dedicated time series dataset was collected at station CB (Bottom depth of 73 m) in the centre of Cambridge Bay (**Figure 3.1b**). Hydrography, nutrient concentrations and phytoplankton biomass were measured through a hole in the sea ice cover 1-2 times a month from February to June. After sea ice melted out of the bay in July, weekly sampling commenced via a 6-m boat through to the end of September. During this period, additional measurements were carried out, including phytoplankton taxonomic composition and *in situ* production estimates. Water sampling depths for the time-series station were 2, 10, 20, 30, 40, and 65-72 m (i.e., bottom sample). Additional to station CB, surface water samples along the wastewater system were collected on six occasions, spanning before and during wastewater discharge into the bay for the examination of particulate organic matter (POM) and inorganic nutrient concentrations. Sampling occurred at the main lagoon, the wetland along the discharge route, and at an outfall location just offshore (**Figure 3.1b**). Additional data including nutrient

concentrations and release flow velocity of the wastewater system was provided by the municipal government.

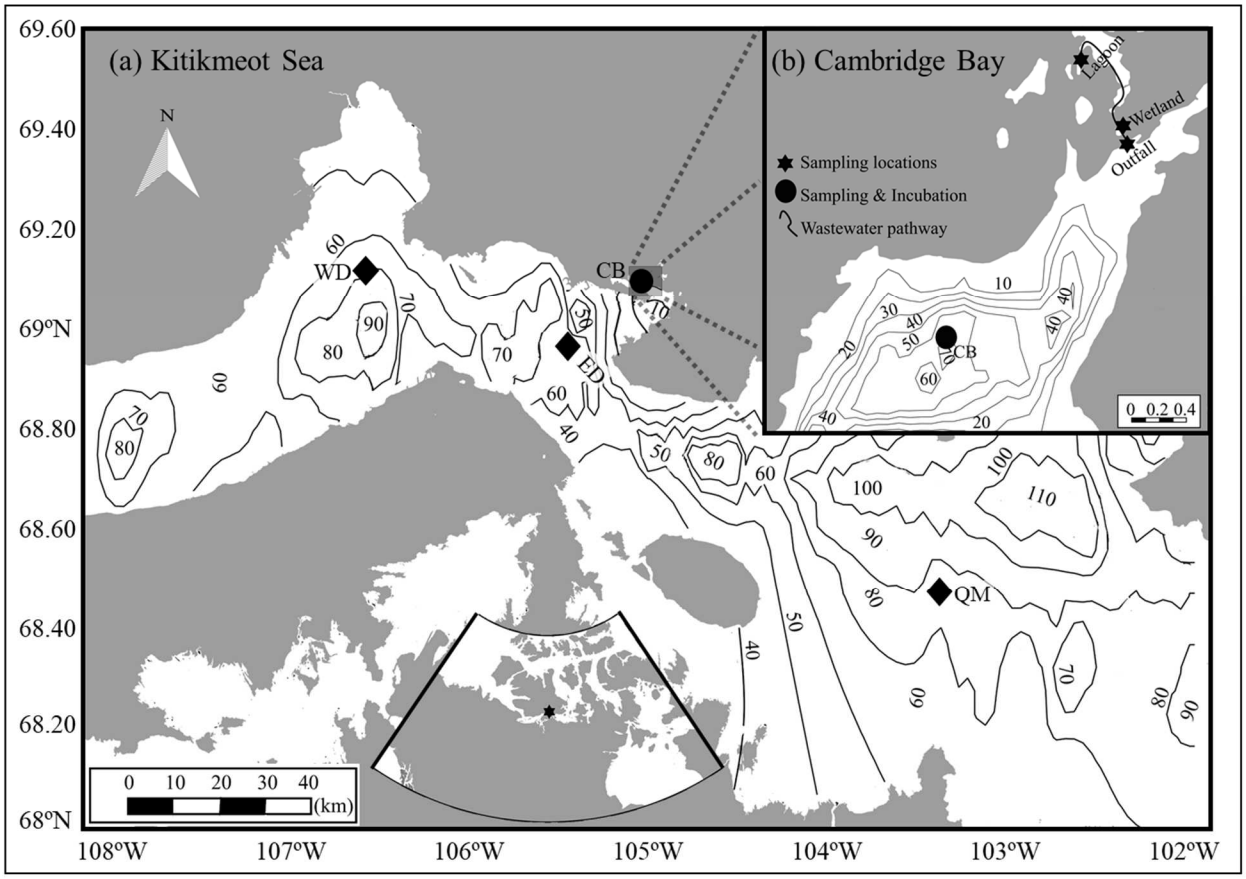

Figure 3.1. Map of study sites in: (a) the Kitikmeot Sea, including West Dease Strait (WD), East Dease Strait (ED), and Queen Maud Gulf (QM), and (b) Cambridge Bay (CB), including wastewater pathway (solid-line illustrates the flow of wastewater) sites, Lagoon, Wetland and Outfall. The Outfall was 5 m downstream (to the right) and 1 m offshore from the Wetland stream mouth.

3.2.2 Hydrography

Distributed profiles of conductivity, temperature, and depth (CTD), photosynthetically active radiation (PAR, 400-700 nm), and *in vivo* chlorophyll *a* concentration were collected using a 19 Plus CTD with a flat spectral PAR sensor (Sea-Bird) and ECO-AFL/FL fluorometer (WET Labs) mounted to a 55 ECO water sampler (Sea-Bird) while onboard the *R/V Martin Bergmann*. Profiles for the CB time-series used a Concerto CTD (RBR Ltd., Canada) with Cyclops fluorometer (Turner) and a spherical 193 PAR sensor (LI-COR). To estimate daily PAR in

support of the *in situ* primary production incubations, the spherical 193 PAR sensor was attached to the incubation rope at a 2-m water depth and logged over each 24-hr incubation period.

3.2.3 Nutrients

Nutrient concentrations were measured using three separate nutrient analyzers due to differences in detection ranges and nutrient analyzed. All seawater samples were collected using Niskin water samplers. Prior to subsampling for nutrients, a syringe, filter holder and 15-mL conical tubes (all acid-washed) were rinsed 3 times with sample. Subsamples were then filtered through pre-combusted (450ºC for 5 hours) 25-mm Whatman GF/F filters into the tubes and stored in the dark at -20°C until sample analysis. Phosphate (PO_4^{3-}), NO_3^-+NO_2^- and silicic acid ($Si(OH)_4$) concentrations in the ocean were measured using a segmented flow analyzer (Seal AutoAnalyzer 3). For determining nutrient concentrations along the wastewater system, surface water samples were collected with an acid-washed and triple sample-rinsed bucket. Subsamples were immediately collected following the abovementioned protocol, including storage in the dark at -20°C until analysis. Wastewater system samples were processed using flow injection analysis (FIA) for NO_3^-+NO_2^- as nitrogen (NO_x-N), NO_2^--N and NH_4^+-N and Hach spectrophotometer (DR 2800, USA) was used for PO_4^{3-} as phosphorus (P). All analyzers determine nutrient concentrations through the colour expressed by the reaction of each nutrient with specific reagents (Giné et al. 1980; Hansen and Koroleff, 1999; Hach Company, 2007). Additionally, some data of NO_x-N and NO_3^--N, which were analyzed at Taiga Environmental Laboratory (www.enr.gov.nt.ca) using Ion Chromatography, in the wastewater system were provided directly from the municipal government overseeing the Cambridge Bay wastewater system. It is important to note that nutrients concentrations are commonly reported in µmol L^{-1} for oceanography and mg L^{-1} for wastewater systems. Therefore, nutrient concentrations from

the wastewater system are indicated in both units.

3.2.4 Phytoplankton Biomass

Chlorophyll *a* (Chl *a*) concentration was used as a proxy of phytoplankton biomass (Falkowski, 1981). Chl *a* concentrations were measured by a fluorometer (Turner Designs Trilogy Laboratory Fluorometer) and via high-performance liquid chromatography (HPLC). To determine Chl *a* concentration in the ocean, 500-mL and 750-mL subsamples for fluorometer and a HPLC analyses, respectively, were filtered through a 25-mm Whatman GF/F filter. In the wastewater system, between 50- and 500-mL subsamples were filtered according to different high particulate concentrations. These filtered samples were kept at -80°C until sample analyses. To determine Chl *a* concentration using the fluorometer, Chl *a* was extracted from each filter in 10 mL of 90% acetone in the dark at 4°C over 20 to 24 h, measured once and then again after acidification with 5% HCl (Parsons et al., 1984). HPLC samples were run at the Isotope Ecology and Environmental Science Laboratory in Hanyang University. Each filter was chopped into small pieces and placed in a conical tube with 1 mL of 100% acetone and 100 µL of internal standard (DHI Water & Environment; canthaxanthin) and then stored in the dark at 4°C for 20 to 24 h. Extracted samples were sonicated for 60 s and centrifuged at 3500 gravity for 5 minutes to incorporate the remaining pigments in each filter into the acetone (Garrido and Roy, 2015). Each sample was filtered through a 0.45-µm polytetra-fluoroethylene syringe filter to remove impurities and then into a HPLC analyzer vial. The analyzer injected 50 µL from each vial into a Waters Symmetry C_8 column for peak separation (Zapata et al., 2000). Phytoplankton pigments were measured using a UV/VIS absorbance detector and chlorophyll-related compounds were redetected using a fluorescence detector (Alou-Font et al., 2013). Pigment absorption peaks provided in Jeffrey et al. (2005) were referenced to identify pigments in the chromatograms. For

HPLC-Chl *a* determination, phaeopigments and total Chl *a* concentration including Chl *a*, divinyl-Chl *a*, Chl *a* isomer and epimer, and chlorophyllids *a* were summed (Babin et al., 2003; Bricaud et al., 2004).

3.2.5 Phytoplankton Taxonomy

Water samples for taxonomic determination using inverted microscopy were collected at 30 m (near pycnocline depth) and bottom depths of station CB on every sampling day, and at the outfall before and during wastewater discharge. Additionally, phytoplankton were collected at station CB via an entire water column vertical plankton net (20-µm mesh, 30-cm diameter mouth) tow accomplished during and after wastewater discharge. A 200-mL sub-sample from water samples and filtered seawater (FSW)-diluted 200-mL sample taken from the net was preserved with 0.4 mL of Lugol's iodine solution (final Lugol concentration of 0.8%). Subsamples of 50 mL were placed in Utermöhl sedimentation chambers (Utermöhl 1958) with cells settling over a 24-h period. Cell enumeration occurred under low and high magnification (100 to 600 times) through an inverted microscope (Nikon, Ti-S) with a minimal cell diameter enumeration limit of 2-5 µm. Phytoplankton were identified to the lowest taxa possible based on Tomas (1997).

3.2.6 Primary Production and Particulate Organic Matter (POM)

In situ primary production incubations were performed throughout the study using stable ^{13}carbon isotope (^{13}C) as a tracer for carbon uptake (Hama et al., 1983). Incubation water depths included 2, 10, 20, 30, and 40 m. Immediately after sample collection and shaded from the sun, three 1000-mL subsamples were spiked with 2.4 mg of ^{13}C from a labelled sodium bicarbonate salt (Sigma-Aldrich) for each water depth (Miller III and Dunton, 2007). Two incubation bottles were positioned along a weighted tether to hang at the water depth they were sampled from for

~24 h. The other incubation bottle was promptly filtered through pre-combusted GF/F filters without incubation in order to make a ^{13}C Time zero (T0) sample. After the incubation time, bottles were retrieved and filtered through pre-combusted GF/F filters. All filtered samples were stored at -80°C until analysis down south. A continuous-flow isotope ratio mass spectrometer (22-20 Stable Isotope Ratio Mass Spectrometer with Europa EA-GSL, SERCON) was used to measure ^{13}carbon atom (At.) and the ^{13}C:^{12}C ratio of particles. Filter processing included drying filters overnight at 60°C, then storage for an additional 24 h in a desiccator saturated with HCl fumes to remove inorganic carbon, and then drying again for another 24 h (JGOFS 19, 1996). Completely dried filter samples were wrapped into pressed tin capsules. For each analysis cycle, four different weights of the standards (Vienna Pee Dee Belemnite) and empty tin capsules as blanks were analyzed with samples to calibrate and confirm instrument stabilization. Particulate organic carbon (POC) and nitrogen (PON) concentrations were calculated using the calibration regression between 4 and 6 points (R>0.99). The following equation (1) from Hama et al. (1983) was used to calculate photosynthetic rates:

$$P = \frac{C}{t} \cdot \frac{(A_{is} - A_{ns})}{(A_{ic} - A_{ns})}, \qquad (1)$$

where P is the photosynthetic rate (mg m^{-3} h^{-1}); C is POC concentration of the incubated sample (µg C L^{-1}); t is the time (h); A_{is} is atomic weight percent of ^{13}C in the incubated sample; A_{ic} is atomic weight percent of ^{13}C in the total inorganic carbon; A_{ns} is atomic weight percent of ^{13}C in the natural environment, which is the T0 sample. To calculate values of A_{ic}, dissolved inorganic carbon (DIC) concentrations in Cambridge Bay were obtained from Duke (2019). Incubation depths (*see section 2.1*) did not usually match the deep chlorophyll maximum (DCM) depth. To incorporate the DCM into the integrated production estimate, a second-order polynomial regression was fit to the assimilation number, the ratio of photosynthetic rate to calibrated *in vivo*

Chl *a* concentration (Valiela, 1995), versus PAR transmittance at the sampling depths. Regressions were accomplished for each day with R^2 always >0.97. This daily incubation relationship was applied to the respective day's profile of PAR transmittance and calibrated *in vivo* Chl *a* fluorescence to estimate primary production profiles that incorporated the DCM and then integrated over 40 m (lowest incubation depth). These estimates are expressed as:

$$\frac{P}{Chl\ a} = aPAR^2 + bPAR + c, \quad (2)$$

$$PE_Z = (aPAR^2 + bPAR + c)_z \times Chl\ a_z, \quad (3)$$

$$PP = \int_{0\ m}^{40\ m} [PE]_z\, dz, \quad (4)$$

where PE_Z is estimated photosynthetic rate (mg m^{-3} h^{-1}) at depth z (m) using formula (2) from a polynomial regression; and PP is daily integrated primary production (mg C m^{-2} d^{-1}).

3.2.7 Statistical Analysis

Least-squares polynomial and linear regressions were applied to some data for analysis steps, as explained above. To test the impact of wastewater, a one-way analysis of variance (ANOVA) was used to compare primary production measured during the discharge versus the one measured before and after discharge and then followed all pairwise multiple comparison procedures (Dunn's Method). All statistical tests were performed in SigmaPlot 12.5.

3.3 Results

3.3.1 Nutrients and Environmental Conditions in the Kitikmeot Sea

Distributed sampling took place between 21 July and 9 August shortly after the local sea-ice break-up date. During this period, below 30 m depth, averaged temperature ranged between -1.5 and -0.5 °C and salinity was observed to decrease westward from Queen Maud Gulf through Dease Strait. However, the highest salinities in the region were observed within Cambridge Bay (**Table 3.1**).

Table 3.1. Averaged salinity and temperature below 30 m depth at distributed stations CB, WD, ED, and QM in the Kitikmeot Sea during early summer of 2013 to 2018.

	Salinity				Temperature (°C)			
Year	CB	WD	ED	QM	CB	WD	ED	QM
2013	29.5	28.7	28.7	29.2	-1.5	-1.3	-1.0	-1.4
2014	29.4			29.2	-1.1			-1.5
2015		28.5	28.7			-1.1	-1.1	
2016	29.6	28.8	29.2	29.4	-1.4	-1.2	-1.3	-1.5
2017	29.5		28.7		-0.9		-0.5	
2018	29.4	28.4	28.4	28.7	-0.6	-1.2	-1.0	-1.3

NO_3^-+NO_2^-, $Si(OH)_4$ and PO_4^{3-} concentrations below a 20-30-m surface pycnocline between 2013 and 2018 ranged from 0 to 4.37, 2.74 to 17.77, and 0.36 to 1.34 µmol L^{-1}, respectively. NO_3^-+NO_2^- concentrations above the pycnocline averaged 0.31±0.28 µmol L^{-1} across the Kitikmeot Sea. Outside of the bay, averaged NO_3^-+NO_2^- concentrations below the pycnocline were 0.85±0.79 in WD, 1.03±0.46 in ED, and 0.93±0.77 µmol L^{-1} in QM (**Figure 3.2a**). In contrast, within CB, NO_3^-+NO_2^- concentration below the pycnocline averaged 2.67±1.30. In particular, NO_3^-+NO_2^- concentrations increased to 4.37 µmol L^{-1} between 2013 and 2015 and then decreased to 1.07 µmol L^{-1} in 2016 and gradually increased to 3.57 µmol L^{-1} again in 2018. This aspect of increase and decrease tended to be similar to temperature change and to be opposite to salinity change (**Table1; Figure 3.2a**). During the 6-year period, NO_3^-+NO_2^-:

$Si(OH)_4$ (N:Si) and $NO_3^-+NO_2^-$: PO_4^{3-} (N:P) molar ratios averaged 1.24±0.68 and 0.17±0.10 outside of the bay, respectively (**Figure 3.2b, c**). In contrast, N:Si and N:P ratios at CB averaged 2.75±1.20 and 0.27±0.12, respectively, equating to 1.6 and 2.2 times greater than those outside the bay, respectively. It is significant to note that in 2016, $NO_3^-+NO_2^-$ concentrations, N:Si and N:P ratios at CB compared closely to those outside the bay.

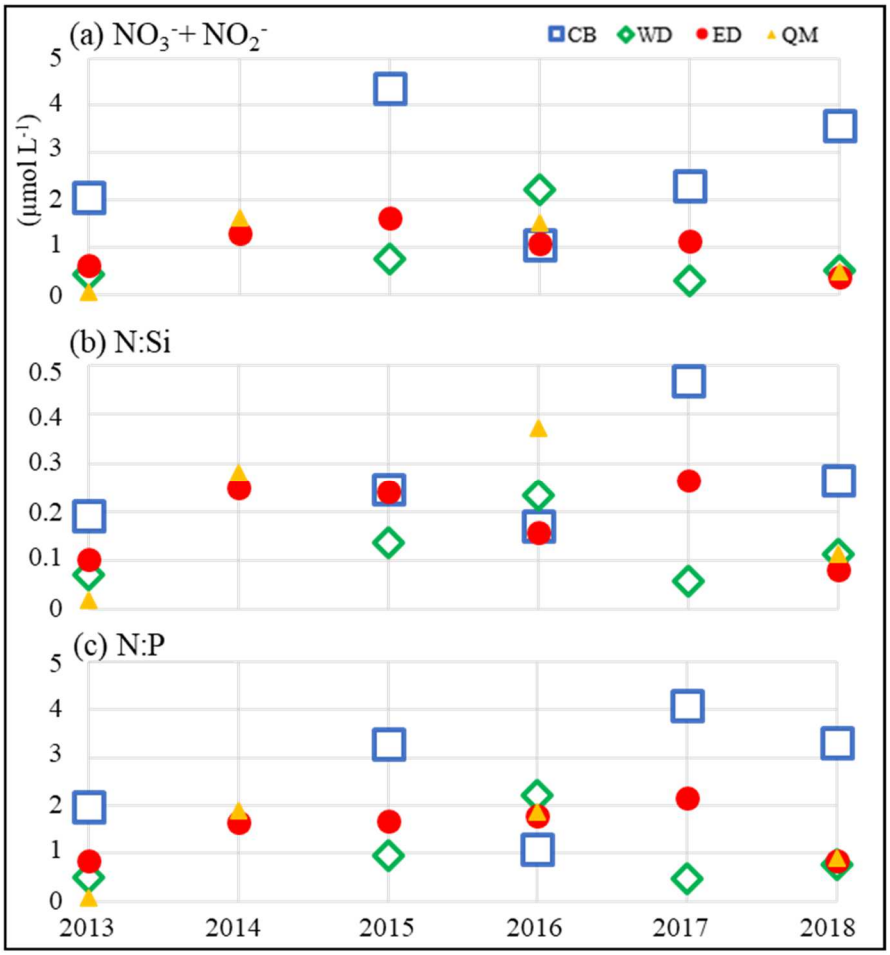

Figure 3.2. Interannual time series of (a) $NO_3^-+NO_2^-$ concentration, (b) $NO_3^-+NO_2^-$: $Si(OH)_4$ (mol:mol; N:Si) and (c) $NO_3^-+NO_2^-$: PO_4^{3-} (mol:mol; N:P) below the pycnocline in the Kitikmeot Sea from 2013 to 2018.

3.3.2 Nutrients and Biotic Parameters at the Wastewater Pathway

Between 23 July and 24 August, a total of 40,404 m^3 (336.7 h of active discharge at 120 m^3 h^{-1}) of wastewater effluent was released. On 16 July before the period of wastewater discharge, NO_x-N, NH_4^+-N and PO_4^{3-}P concentrations at the outfall were 0.46, 15.19 and 0.86 µmol L^{-1}, respectively (**Table 3.2**). During the period of the discharge, wastewater effluent nutrient concentrations ranged from 8.35 (NO_x-N), 159.10 (NH_4^+-N) and 24.05 µmol L^{-1} (PO_4^{3-}P) on 28 July to 51.08 (NO_x-N), 5.99 (NH_4^+-N) and 4.21 µmol L^{-1} (PO_4^{3-}P) on 23 August. Concentrations of PON and POC in the wetland ranged between 4.03 and 4.10 mg N L^{-1} and 24.14 and 27.55 mg C L^{-1}, respectively. Chl *a* concentration at the outfall increased from 4.07 µg L^{-1} before discharge to an average of 40.05 ± 17.53 µg L^{-1} during the discharge period (**Table 3.2**).

Table 3.2. Nitrogen and phosphorous compound, POM and Chl *a* concentrations in the wastewater pathway before and during the discharge. LA, lagoon; WET, wetland; OUT, outfall; SAT, Saturday (no discharge); EVE, evening (weak flow of wastewater effluent).

	Date	Area	Nitrogen and Phosphorous µmol L^{-1} (mg L^{-1})					POM mg L^{-1}		µg L^{-1}
			NO_x-N	NO_3^--N	NO_2^--N	NH_4^+-N	PO_4^{3-}-P	PON	POC	Chl *a*
Before	7/5*	LA		7.58 (0.47)						
		WET		1.61 (0.10)						
		OUT		5.81 (0.36)						
	7/16	LA	1.89 (0.09)		2.03 (0.09)	571.00 (10.30)	42.54 (4.04)			
		WET	0.14 (0.01)		0.03 (0.00)	9.37 (0.17)	6.55 (0.62)	0.11	0.97	
		OUT	0.46 (0.02)		0.45 (0.02)	15.19 (0.27)	0.86 (0.08)	0.04	0.36	4.01
During	7/28 (SAT)	LA	28.21 (1.30)		31.95 (1.47)	708.48 (12.78)	41.44 (3.94)			
		WET	8.35 (0.38)		8.52 (0.39)	159.10 (2.87)	24.05 (2.28)	4.03	24.14	
		OUT	1.03 (0.05)		0.78 (0.04)	18.29 (0.33)	0.51 (0.05)	0.18	1.19	27.65
	7/30*	LA								
		WET		11.13 (0.69)						
		OUT		10.81 (0.67)						
	8/23* (DAY)	LA	57.82 (2.66)							
		WET	51.73 (2.38)							
		OUT	48.47 (2.23)							
	8/23 (EVE)	LA								
		WET	51.08 (2.35)		18.82 (0.87)	5.99 (0.11)	4.21 (0.40)	4.10	27.55	
		OUT	2.52 (0.12)		2.46 (0.11)	13.30 (0.24)	1.84 (0.18)			52.44

* data from these dates were provided by the municipal government

At the outfall and before wastewater discharge on 16 July, fucoxanthin accounted for 67% of the total accessory pigments (**Figure 3.3a**). This percentage compared closely with an observed 64% of unidentified nanoflagellates (3-10 µm) in the enumerated phytoplankton taxonomic composition from the same sample (**Figure 3.4a**). After discharge commenced, the relative contributions of lutein and Chlorophyll *b* (Chl *b*) to total accessory pigments increased to

41 and 49% on 28 July and 53 and 30% on 23 August, respectively. Similarly, there was a shift in taxonomic composition to a dominance of Chlorophyceae (71%; mainly including *Ankistrodesmus setigerus*, *Dictyosphaerium sp*. and *Scenedesmus acuminatus*) and Chrysophyceae taxa (20%; especially *Dinobryon balticum*) on 28 July.

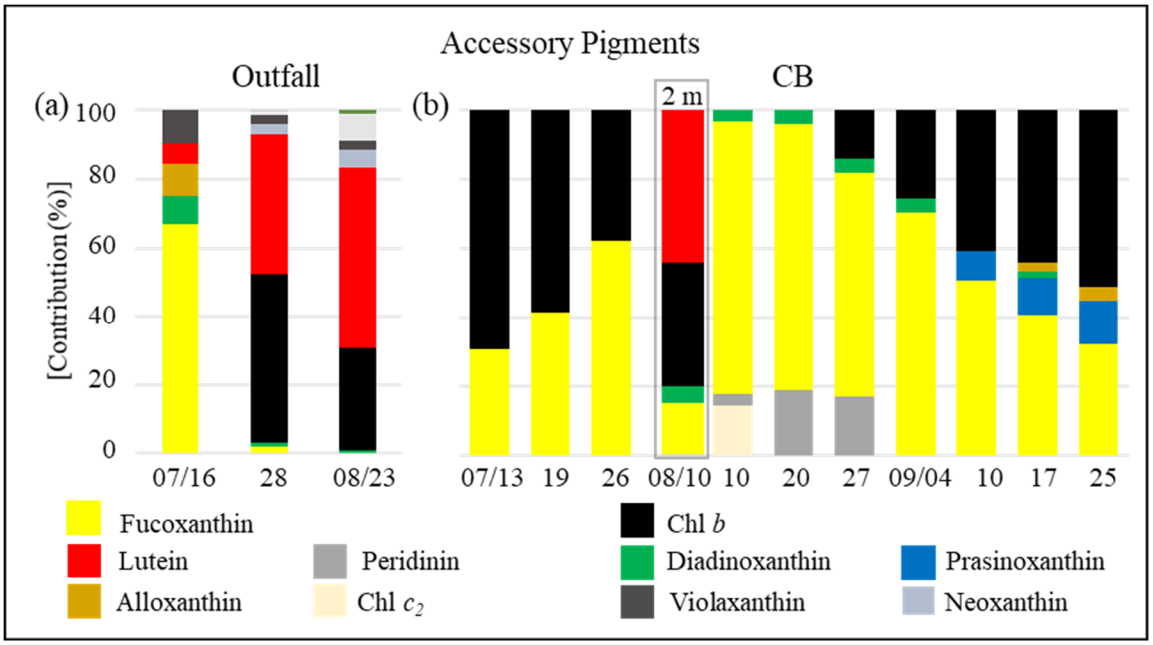

Figure 3.3. Bar charts showing the relative contribution (%) of accessory pigments (a) at the outfall and (b) near DCM depths (the sampling depth: 30 or 40 m) and at 2 m depth on 10 August in CB.

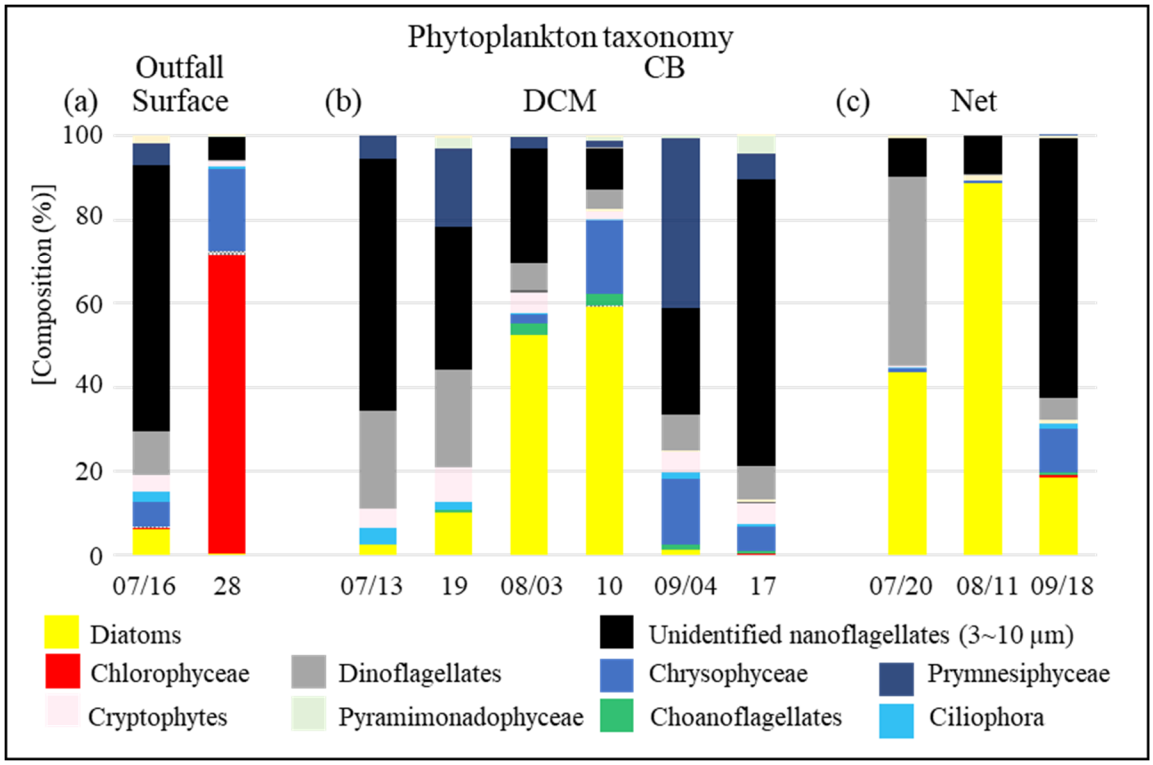

Figure 3.4. Bar charts showing the percentage of phytoplankton taxonomic composition (a) at the outfall, (b) near DCM depths (the sampling depth: 30 or 40 m) in CB, and (c) the taxonomic composition for whole depth in CB before, during and after the discharge depth.

3.3.3 Time series of Cambridge Bay (CB) in 2018

3.3.3.1 Hydrography

Evident in the Brunt-Vaisala frequency and controlled by a halocline, a persistent pycnocline at a salinity of 29 and a temperature of -0.5°C was observed at CB between 20 and 30 m throughout the study (**Figure 3.5**; black dashed line). Above the pycnocline, salinities decreased over the first half of the study with ice melt and finally break-up on 9 July 2018 (as seen by NASA Worldview), forcing a seasonal surface halocline that only mixed down to the depth of the persistent pycnocline towards the end of the study in late September. Surface temperature was near freezing at -1.56°C above the pycnocline while sea ice covered the bay, then rapidly increased to >5°C by ice break-up. Below the pycnocline, salinity and temperature were relatively stable, averaging 29.4 and -0.7, respectively (**Figure 3.5**). The euphotic zone,

determined by the depth of 1% PAR transmittance, reached below 60 m following ice break-up and then shoaled to range between 30 and 50 m by late July (**Figure 3.5**; white dashed line).

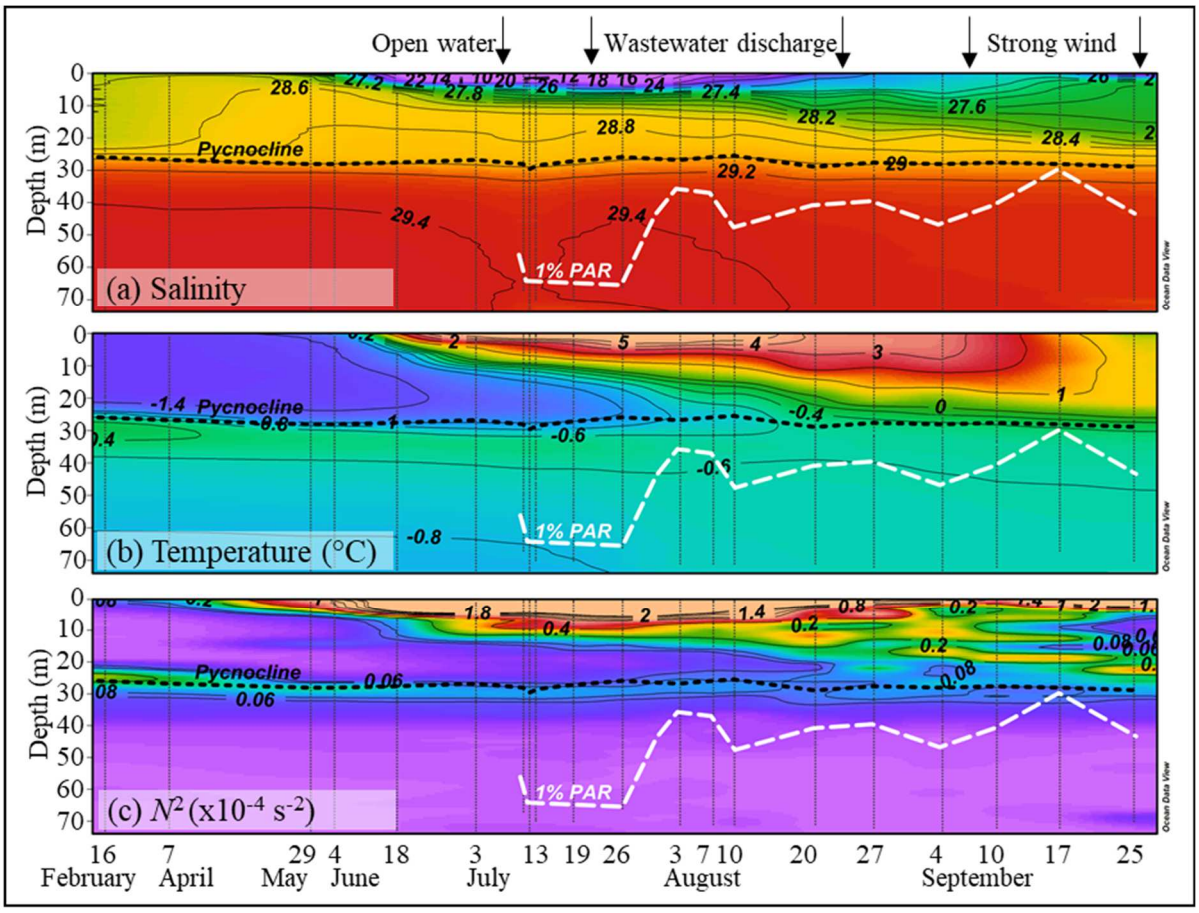

Figure 3.5. Time-series of (a) salinity, (b) temperature and (c) Brunt-Vaisala frequency from February to September at the Cambridge Bay site (CB); the black dashed line represents the depth of the persistent pycnocline (halocline) at a salinity of 29 and a white dashed line represents the depth of 1% PAR transmittance.

3.3.3.2 Nutrients and Phytoplankton Biomass

Over the study, NO_3^-+NO_2^-, $Si(OH)_4$ and PO_4^{3-} concentrations averaged 3.86 ± 2.13, 13.63 ± 6.83 and 1.06 ± 0.32 µmol L^{-1} below the persistent pycnocline, respectively (**Figure 3.6a-c**). In contrast, surface NO_3^-+NO_2^-, $Si(OH)_4$ and PO_4^{3-} concentrations were much lower throughout the study, averaging 0.37 ± 0.31, 4.02 ± 1.79 and 0.56 ± 0.12 µmol L^{-1}, respectively. NO_3^-+NO_2^- above the pycnocline decreased to <0.5 µmol L^{-1} by 13 July and remained depleted

thereafter. Surface $Si(OH)_4$ and PO_4^{3-} concentrations also decreased during this initial period. Interestingly, $Si(OH)_4$ concentrations further decreased between 19 and 26 July, which was not observed in $NO_3^-+NO_2^-$ concentrations. However, both $NO_3^-+NO_2^-$ and $Si(OH)_4$ decreased below the pycnocline by 20 August. It is significant to note that PO_4^{3-} concentrations decoupled from trends of the other inorganic nutrients, tending to increase on average during the second half of the study. Surface Chl *a* concentrations started to increase immediately under the ice during late April to early May, deepening in its increase as ice break-up occurred on 9 July and slightly increasing until 19 July resulting in a distinct DCM (**Figure 3.6d**). During this period, areal Chl *a* concentrations increased from 2.39 to 27.69 mg Chl *a* m^{-2}. Following the commencement of wastewater discharge on 23 July, Chl *a* concentration rapidly increased while the DCM depth increased to below the pycnocline, but within the euphotic zone. Areal Chl *a* concentration during the period of wastewater discharge averaged 58.54 ± 28.10 mg Chl *a* m^{-2}. Maximum Chl *a* concentrations at the DCM averaged 11.13 ± 5.87 µg Chl *a* L^{-1} during the period, peaking at 15 µg Chl *a* L^{-1} at 25 m on 10 August. After wastewater discharge ended on 23 August, Chl *a* concentration stayed at a relatively high level until 27 August (42.55 mg Chl *a* m^{-2}) and then rapidly decreased to 19.57 mg Chl *a* m^{-2} on 4 September.

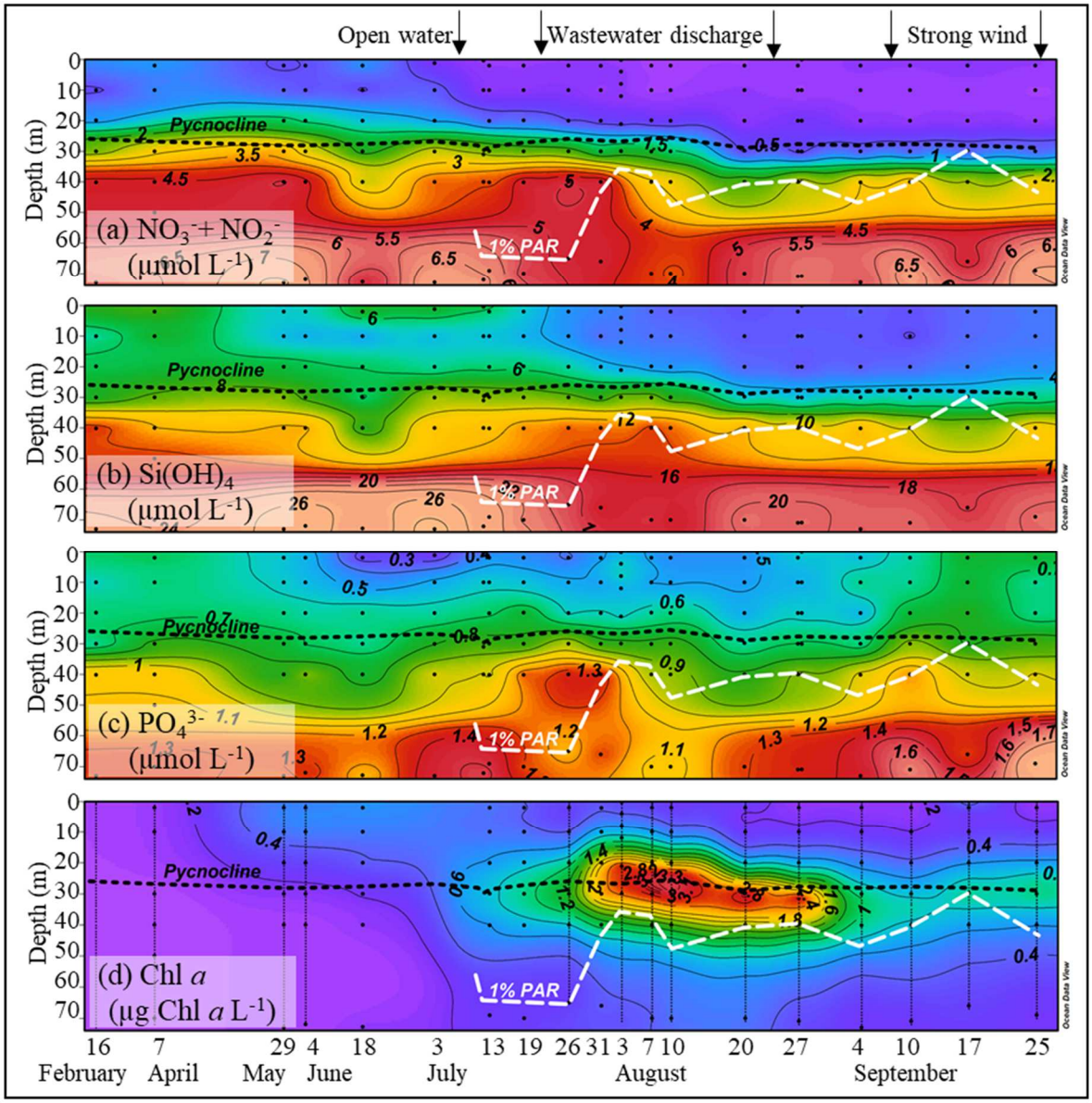

Figure 3.6. Time-series of (a) $NO_3^-+NO_2^-$, (b) $Si(OH)_4$, (c) PO_4^{3-} and (d) Chl *a* concentration; big vertical dots on the graphs are the sampling depths, and vertical dashed lines on (d) represent CTD cast data calibrated to water sample Chl *a*; black and while bold-dashed lines on each graph are the depths of the persistent pycnocline (halocline) and 1% PAR, respectively.

Before wastewater discharge commenced, PON and POC concentrations ranged from 4.22 to 16.79 µg N L^{-1} and from 35.12 to 118.05 µg C L^{-1}, respectively (**Figure 3.7a,b**). During this period, POC:PON and POC:Chl *a* ratios averaged 7.58 ± 1.05 and 113.95 ± 46.87, respectively (**Figure 3.7c,d**). During the period of wastewater discharge, PON and POC concentrations averaged 13.90 ± 3.27 µg N L^{-1} and 114.22 ± 27.99 µg C L^{-1}, respectively, while

the POC:PON ratio above the persistent pycnocline averaged 9.01 ± 1.74. Similarly, POC:Chl *a* ratio above the pycnocline increased from an averaged 140.97 ± 41.28 to 887.70 ± 998.28 during wastewater discharge. Both POC:PON and POC:Chl *a* ratios peaked at the surface on 3 August, reaching 13.6 and 4030, respectively. During the discharge period near the pycnocline at 30 m, POC:PON and POC:Chl *a* ratios averaged 7.12 and 49.3, respectively. After the end of wastewater discharge, peak PON and POC concentrations deepened to occur below the pycnocline and slowly decreased throughout the water column towards the end of the study. Ratios of POC:PON and POC:Chl *a* also tended to decrease during September, but remained relatively higher in surface waters.

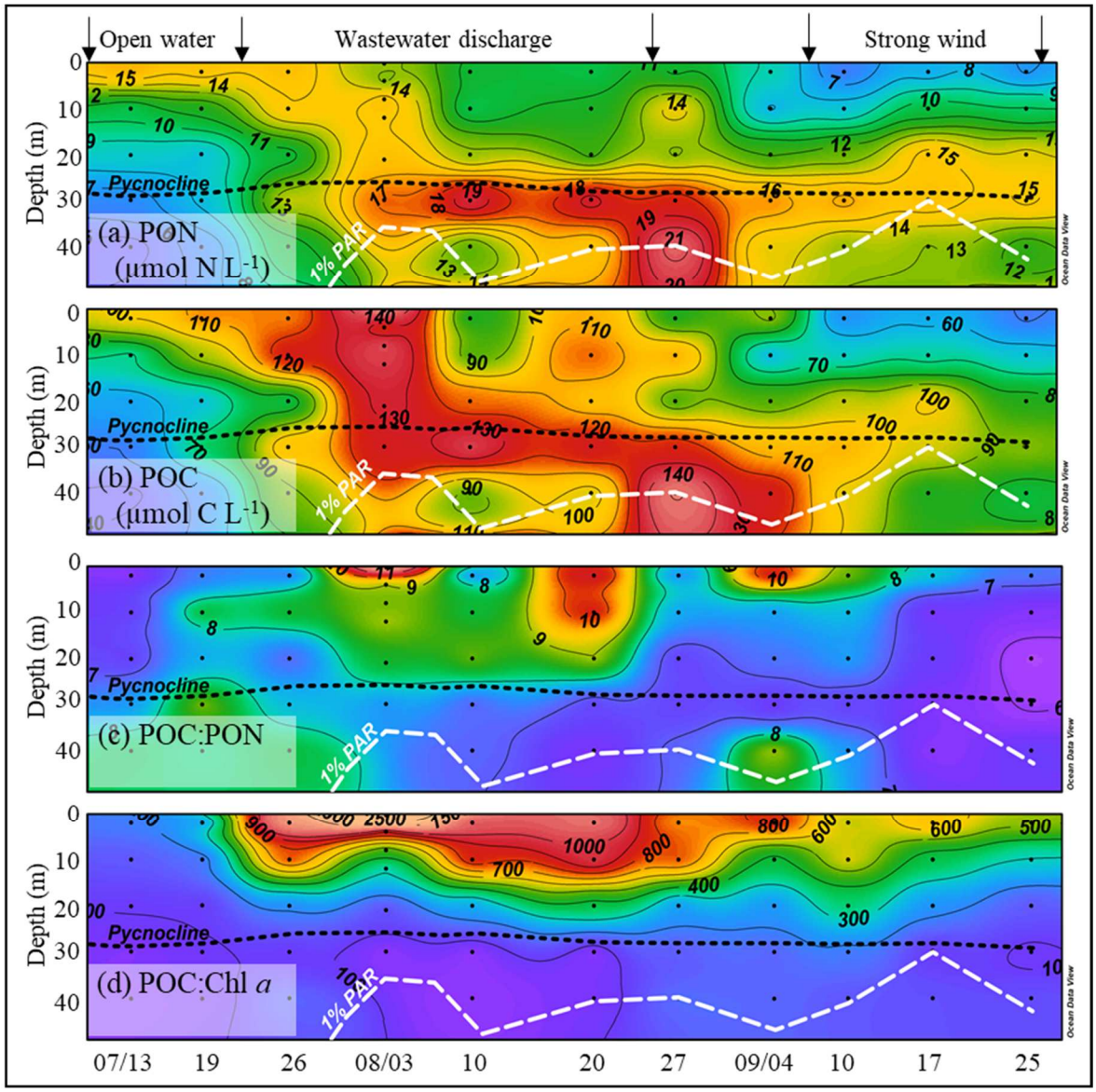

Figure 3.7. Time-series of (a) PON and (b) POC concentrations and the ratios of (c) POC:PON and of (d) POC:Chl *a* at CB from July to September 2018.

3.3.3.3 Phytoplankton Pigment and Taxonomic Composition

Before wastewater discharge commenced, Chl *b* and fucoxanthin near the DCM contributed an averaged 64 and 36% to all accessory pigments, respectively, with nanoflagellates making up the dominant taxa followed by dinoflagellates (**Figured 3.3b, 3.4b**). The water column-integrated vertical phytoplankton net tow collected during the same period on 20 July, revealed a strong presence of diatoms, with dinoflagellates, diatoms and nanoflagellates

accounting for 45, 44, and 9% of the total phytoplankton composition, respectively (**Figure 3.4c)**. After wastewater discharge commenced, the contribution of fucoxanthin and diatoms (dominated by centric diatoms, *Attheya septentrionalis*, *Thalassiosira* sp., and *Chaetoceros* spp.) increased to >60% near the DCM and then greatly decreased following termination of wastewater discharge. In contrast, Chl *b* and nanoflagellate contributions were observed to increase during this later period. It is significant to note the appearance of a strong lutein contribution to accessory pigments at 2 m at CB on 10 August. Chlorophyceae *Scenedesmus acuminatus* and Chrysophyceae *Dinobryon balticum* also emerged at CB station, albeit at <1% contribution to the community composition, at 30 m on 10 August and within the vertical net tow on 11 August (**Figure 4c**).

3.3.3.4 Primary Production

After sea ice completely melted and before the start of wastewater discharge, integrated daily primary production was low, averaging 40.99 ± 2.87 mg C m^{-2} d^{-1} (**Figure 3.8b**). After discharge commenced, integrated daily primary production increased by an order of magnitude, peaking on 3 August at 454.47 mg C m^{-2} d^{-1}, with production focused at the DCM. Following this peak, yet still during the discharge period, primary production decreased to approximately 100 mg C m^{-2} d^{-1} (**Figure 3.8b**). Then, after discharge was stopped, integrated daily primary production decreased to 33.19 mg C m^{-2} d^{-1} on 4 September. However, coincident with strong winds, a slight increase in production was observed at the surface on 10 and 17 September with an integrated daily rate of 63.41 ± 4.99 mg C m^{-2} d^{-1}.

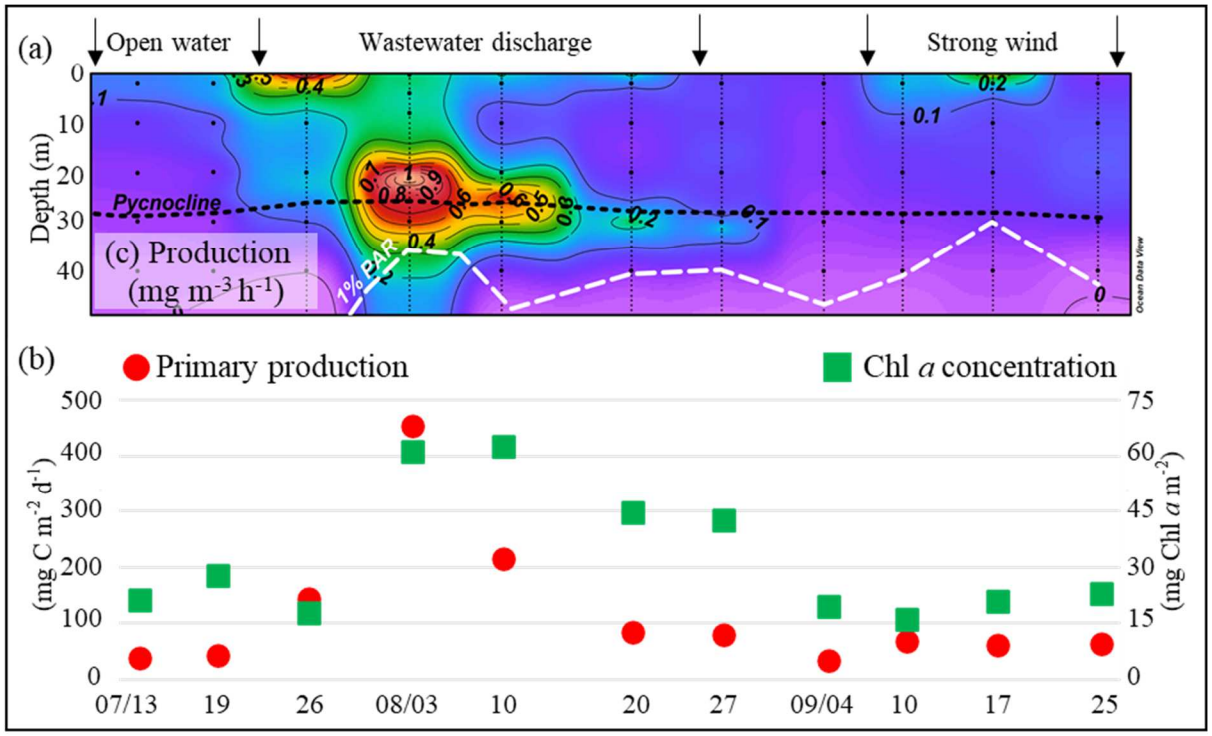

Figure 3.8 Time-series of (a) estimated production rates at CB from July to September 2018 (3 August data from *R/V Bergmann voyage* where water samples were taken within euphotic depths), and (b) 40-m integrated primary production (red dots) and Chl *a* concentrations (green squares).

As a result of an ANOVA and comparison test (Dunn's Method), primary productions during the period of wastewater water discharge were significantly different (*p-value* = 0.006) from those from the periods before and after the discharge.

3.4 Discussion

3.4.1 Hydrography and Nutrients in the Kitikmeot Sea

The Kitikmeot Sea is defined as nitrogen-deplete, strongly limiting primary production in the region (Campbell et al. 2016; Dalman et al. 2019; Kim et al. *Submitted*). Slight decreases in salinity (**Table 3.1**) from Queen Maud Gulf (QM) toward Dease Strait (ED and WD) provide evidence that saltier waters mainly enter the region from the east, slightly mixed as they transit across the narrow and shallow Victoria Strait to the east of QM. These saltier waters likely originate from the Canadian Beaufort Sea and to a lesser extent the Canada Basin, both classified as oligotrophic nitrogen-deplete surface waters (Li et al. 1999; Ardyna et al. 2011), following the coastal boundary current (coast to right of flow direction) around Victoria Island. However, high riverine input to the Kitikmeot Sea and the associated low salinities set up a local peak in steric height in this southwestern portion of the Northwest Passage, slowing circulation through the region (McLaughlin et al., 2004). The low nitrogen input and stagnant circulation set up some of the most nitrogen-deplete waters of the Arctic Ocean. Based on Redfield stoichiometry associated with the phytoplankton assemblage, the ratios of 16N:15Si:1P are broadly used as the standard for the nutrient ratio for all oceans (Brzezinski, 1985). Averaged nutrient ratios in the Kitikmeot Sea (not including CB) for this study were 1.1N:7Si:1P (**Figure 3.2b, c**), implying severe nitrogen-limitation for the sea because the N:P ratio is much less than the Redfield ratio of 16N:15Si:1P (Brzezinski, 1985). In particular, it is less than 5, which signifies nitrogen-limitation of the physiological range of N:P in phytoplankton (Geider and La Roche, 2002). The relatively higher concentrations of $Si(OH)_4$ suggest a potential for the prevalence of diatom production (Yool and Tyrrell, 2003) in spring when inorganic nitrogen compounds are at their greatest annually due to winter mixing and remineralization processes (Tremblay et al., 2015).

The hydrography of CB and its nutrient budget are somewhat different from the rest of the Kitikmeot Sea. A fjordal-type bathymetry with shallow sills at 20 m encompassing the inner bay and 11 m sill at the outer edge of the bay, and a relatively deep basin of >70 m provides potential for trapping dense water formed during sea-ice formation (**Figure 3.1**; Gade et al., 1974). Indeed, the persistent pycnocline and saltier waters of CB (**Figure 3.5**), as well as NO_3^- +NO_2^- concentrations greater on average than those in Dease Strait (**Figure 3.2a**), characterize the bay as a semi-enclosed system. Furthermore, the greater than average N:Si and N:P ratios within the bay suggest an additional N-source to the bay. However, interannual fluctuations in NO_3^-+NO_2^- concentrations and N:Si and N:P ratios in the bay, which equated to those outside of the bay in 2016 (**Figure 3.2**), suggest periodic ventilation of the bay with external waters from Dease Strait. The colder and more saline waters observed below the pycnocline in 2016 (as well as 2013), suggest a link of ventilation to sea ice formation and associated brine rejection (**Table 3.1**).

3.4.2 Nitrogen Dynamics and Biological Activities in the Wastewater Treatment System

Results clearly demonstrate an input of high-nitrogen compounds into the bay from the local wastewater system. However, the type of nitrogen compound varied during discharge. Remineralization of organic matter, algal uptake and growth, and nitrification of NH_4^+ to NO_3^- (Daims et al., 2006; Delgadillo-Mirquez et al., 2016) were active processes in the wastewater system, evidenced by increases in Chl *a*, POC and PON concentrations, decreases of initially high NH_4^+-N and PO_4^{3-}P concentrations and increases in NO_3^--N concentrations during the discharge period (**Table 3.2**).

Prior to the start of discharge on 16 July, unidentified nanoflagellates were dominant at the outfall (**Figure 3.4**). The high fucoxanthin composition and presence of violoxanthin, yet

lack of diatoms at the site suggests that the nanoflagellate was likely a chrysophyte (i.e., golden-brown flagellate). The taxa could be a potential remnant community of the ice melt, known to support a relatively abundant composition of freshwater-brackish chrysophytes such as *Dinobryon* spp. (Bursa, 1963; Gradinger, 1996; Mundy et al., 2011), or, more simply, a native taxa of the estuary. During discharge on 28 July, lutein-rich chlorophytes dominated the outfall algal community. The appearance of this community associated with the rapid increase in Chl *a* concentration at the outfall suggests an origin from the wastewater system, which was the likely community drawing down a portion of the NH_4^+-N and PO_4^{3-}P during the discharge period.

3.4.3 Wastewater Effluent and its Impact on Phytoplankton Production

Based on average values and the total amount of wastewater effluent, we estimate 6500 mol of NH_4^+-N, 2100 mol of NO_x-N, 170 kg N of PON and 1100 kg C of POC were released from the wastewater system into receiving waters of the bay in 2018. Regardless of the N-compound source, wastewater release had a significant impact on primary production in the bay. Similar production estimates between 13 and 19 July, combined with the depletion of surface NO_3^-+NO_2^- to <0.5 µmol L^{-1} and increase in the POC:PON and POC:Chl *a* ratios above the pycnocline suggests onset of N-limitation at CB just prior to the commencement of wastewater discharge. Then on 26 July, right after wastewater discharge started, primary production increased at the surface. Interestingly, the surface production increase was short-lived, with an observed peak in production centred at the pycnocline by 3 August (**Figure 3.8b**). This downward shift in production was associated with a rapid increase in POC:PON and POC:Chl *a* ratios above the DCM and particularly at the surface. This relative increase in carbon content of the POM suggests a shift towards a more heterotrophic community with a likely increase in microbial activity at the surface (Lee et al., 2004; Buchan et al., 2014). The strong lutein

contribution to accessory pigments observed on 10 August at CB in the surface 2 m sample also provides strong evidence that at least a portion of the wastewater chlorophyte-dominated community and associated effluent drifted out to the centre of the bay. Here, they contributed to the increase in POC, but interestingly did not act to accordingly increase PON or Chl *a* concentrations at the surface.

It was at the DCM where production was greatest during wastewater discharge, Chl *a* concentrations peaked at 15.0 μg L^{-1} on 10 August, and POC:PON and POC:Chl *a* (g:g) ratios decreased to 7.12 and 49.3, respectively. These observations suggest the wastewater effluent impact on primary production was concentrated near the DCM in the bay. Indeed, the freshwater chlorophytes *Scenedesmus acuminatus* species that dominated the wastewater effluent and outfall site during wastewater discharge appeared at low concentrations near the DCM on 10 August. It is possible that the wastewater chlorophyte community could have rapidly settled out of the water column near the outfall. Due to the relatively shallow slope of 10 to 40 m near the outfall and the persistent pycnocline at 29 m (**Figure 3.3**), these settled algae and associated effluent particulate matter could have a residence time along the pycnocline. It is possible that cell death and lysis of the settling freshwater community near the pycnocline and resultant release of labile organic matter—influenced by osmotic stress, sloppy feeding by zooplankton, and viral infection (Bronk and Steinberg, 2008)—could rapidly provide accessible N-compounds to the phytoplankton community. Such a mechanism can help explain the restriction of peak primary production at the pycnocline during the wastewater discharge.

Summing up the impact of wastewater effluent, we compared the amount of measured primary production to the total estimated nitrogen input via the wastewater system. During wastewater discharge between 23 July to 24 August, a total primary production of 7.19 g C m^{-2} was

estimated at CB. This value was 68% of the total production at 10.5 g C m^{-2} estimated for the entire study from 9 July to 10 October. Averaging production before and after wastewater discharge and applying it to the wastewater discharge period, we estimated. 1.75 g C m^{-2}. This rate was assumed to represent baseline primary production for the system between 23 July and 24 August, dependent on the available nitrogen in the surface mixed layer. We subtracted this value from 7.19 g C m^{-2} to estimate the wastewater-associated impact on the system to be 5.44 g C m^{-2}. We further compared this value to the nitrogen input estimated from the wastewater effluent. The total contribution from all wastewater N sources converted to C-production using the average POC:PON ratio of 7.12 at 30 m during the discharge period equates to 2,680 kg C. To define the areal extent for this C-production, we used the 20- and 30-m isobaths that connected the outfall and CB sites, justified by the majority of production observed at the DCM below 20 m. The estimate ranged from 1.30 to 1.71 g C m^{-2}, which equates to 24-31% of the overall wastewater production impact (i.e., 5.44 g C m^{-2}). Clearly, the wastewater N input did not fully explain the additional production during the wastewater period. It is noted that toward the end of the discharge period, the DCM and production peak dropped below the persistent pycnocline, which provided the additional N-source to explain the rest of the observed production. Applying our knowledge that the system is semi-enclosed, with little exchange of waters outside of the bay due to the 11-m sill, we estimated total new production associated with the NO_3^-+NO_2^- drawdown at CB. That is, the NO_3^-+NO_2^- concentration profile (0-50 m) measured on 16 February was subtracted from that measured on 25 September. The difference was then integrated from 0-50 m and converted to C production using the PON: POC ratio at 30 m (7.12) to estimate a total NO_3^-+NO_2^--based production of 7.54 g C m^{-2}. Therefore, 72% of total measured production in the bay (10.5 g C m^{-2}) was explained by NO_3^-+NO_2^- drawdown

over the season. Including the production estimate from the wastewater-nitrogen addition. This leaves, 12~16% of production missing from the budget, which was likely supplied from other N-sources such as NH_4^+, which was not measured in the oceanic environment in this study. Indeed, Kim et al. (*In Review*) observed ~1 µmol L^{-1} of NH_4^+ in surface waters during spring under the ice cover, approximately one-third that of NO_3^-+NO_2^- concentrations measured in study from a nearby location in Dease Strait of the Kitikmeot Sea.

3.4.4 Phytoplankton Succession and its Role in Vertical Export of Nitrogen

Changes in phytoplankton taxonomic composition before, during and after wastewater discharge were observed in the bay. Prior to wastewater discharge, the DCM phytoplankton community was dominated by small unidentified nanoflagellates (3-10 µm) (**Figure 3.4b**). With the supply of wastewater effluent N-compounds and the presence of relatively high $Si(OH)_4$ concentrations, the resultant bloom was dominated by diatoms, further reducing surface $Si(OH)_4$ concentrations during wastewater discharge. The dense opal frustule and large size of diatoms can greatly increase their potential to sink (Legendre, 1990; Ragueneau et al., 2000), which helps explain the increased drawdown of NO_3^-+NO_2^- below the persistent pycnocline up to the euphotic depth after the diatom-dominated community began to bloom. As noted in the last section, the additional N-source below the pycnocline in the bay can explain a large portion of primary production not accounted for by wastewater effluent directly. It is suggested that the diatom bloom was required to access this additional N-source. Furthermore, it is through this migration of the bloom to below the pycnocline that N-compounds from the wastewater effluent can be transported to depth and eventually build up as they remineralize to inorganic nitrogen compounds through activities of microorganisms overtime (Garber, 1984; Buchan et al., 2014). This vertical export helps explain the additional N-source suggested for CB derived from the

observed elevated N:Si and N:P ratios in the bay relative to the surrounding Kitikmeot Sea. After wastewater discharge was stopped, the diatom population declined, replaced by smaller flagellate cells, more characteristic of a nutrient-limited post bloom state (Officer and Ryther, 1980; Michael and Glibert, 2000).

Long after wastewater discharge had stopped and surface nutrients were depleted in mid-September, surface production rates slightly increased, coupled with an increase in areal Chl *a* concentrations. This small fall bloom was actually a function surface mixing due to strong winds (wind gusts exceeding 50 km h^{-1} during the period; http://climate.weather.go.ca). To our knowledge, this observation represents one of the first *in situ* documentations of a wind-induced fall bloom in the Arctic. With a later freeze-up and thus longer exposure of surface waters into fall, as well as overall increased windiness of the Arctic, fall phytoplankton blooms have been observed via satellite throughout the Arctic Ocean (Ardyna et al., 2014).

3.5 Conclusion

The Kitikmeot Sea is among the most N-deplete system in the Arctic Ocean. Our findings demonstrate an important local influence of wastewater effluent on primary production in Cambridge Bay. The fjordal-type bathymetry and persistent pycnocline below the exit sill of the bay act to trap much of the wastewater N at depth, causing the marine environment to be unique from that outside of the bay. Evidence from N:P and N:Si inorganic nutrient ratios suggest that the bay can be ventilated; however the cause and timing of this ventilation is speculative at best. N-addition to the system is still relatively minor as the system cannot be considered eutrophic with a maximum production of only 10.5 g C m^{-2} over the open water period with daylight. For example, other regions in the Arctic Ocean have reported phytoplankton production ranging from annual estimate of 56 g C m^{-2} y^{-1} near Resolute Bay, Canada (Welch et al. 1992) to 32.5 g C m^{-2} accumulation over a single bloom in the Chukchi Sea (Arrigo et al. 2012). Furthermore, harmful algal species typically associated with wastewater-influenced blooms, such as *Psuedo-nitzschia spp.* (Pan and Rao, 1997), were not detected at CB during the wastewater discharge. These results suggest that the present impact of wastewater discharge into Cambridge Bay is not critical, but does pose a concern to the region. With the growing population of Canada's Arctic, a monitoring program, and additional research, e.g., to help answer some of the questions raised here, is strongly recommended to improve future management decisions regarding regulation and operation of wastewater systems in Arctic regions.

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CHAPTER FOUR: CONCLUSIONS AND RECOMMENDATIONS

4.1 Summary

Chapter one introduces the thesis topic, explains the underlying importance of the research, states three research objectives, and describes the structure of this dissertation, including contents of the four chapters. Chapter two provides pertinent background information related to the thesis, including descriptions of general processes and concepts including application of this information to understanding the current state and changes in the Arctic region. Effort is placed to describe limiting and influencing factors for phytoplankton growth, with an emphasis on light availability and nutrient dynamics and their environmental controls. Additionally, information on phytoplankton species composition and primary production to explain the interaction among the environmental factors, phytoplankton species composition and primary production is provided. Lastly, a closer examination of wastewater treatment systems and logistical challenges faced in the Arctic environment is discussed. This examination focuses on the thesis case study site of Cambridge Bay, Nunavut, Canada within the Kitikmeot Sea.

Chapter three includes a manuscript prepared for submission to an international journal that examines the effects of wastewater effluent on local primary production in Cambridge Bay as a case study for the Canadian coastal Arctic. The research compares environmental characteristics of the bay to the adjacent Kitikmeot Sea and describes changes in phytoplankton species composition as well as increases in phytoplankton biomass and associated primary production before, during, and after wastewater discharge into the bay. The fjordal-type bathymetry of Cambridge Bay was found to be a semi-enclosed system with a persistent pycnocline centred at 29 m, below the bay's exit sills at 20 and 11 m. The pycnocline and bathymetry led to trapping of organic matter at depth in the system, which resulted in an elevated

nitrogen inventory in the bay relative to the surrounding Kitikmeot Sea. Interannual variability in nutrient concentrations and ratios suggest the bay periodically ventilates its deep waters, although the process driving this occurrence requires further investigation. Within Cambridge Bay and prior to discharge, phytoplankton production was steady and low, and it appeared to use up the surface nitrate inventory by mid-July, supported by nutrient limitation onset signals of high POC:PON and POC:Chl *a* ratios and a dominant nanoflagellate community. With the onset of wastewater release in late July, phytoplankton production rapidly increased with large diatoms dominating the community composition. It was estimated that wastewater effluent released 6500 mol of NH_4^+-N, 2100 mol of NO_x-N, and 0.2 kg N of PON into the marine system during discharge in 2018. This nitrogen addition accounted for up to 31% of production above the bay's background levels (i.e., averaged rate before and after wastewater release). The additional nitrogen stored at depth in the bay was able to explain the additional production, which is believed to derive from the concentration of organic matter originating from the wastewater system, but trapped at depth. Although the system is concluded to not yet be eutrophic, caution is raised at the rapid response of the marine system to the wastewater release with a strong recommendation for establishment of a future monitoring and research program for the bay.

4.2 Conclusions and Recommendations

Ongoing global warming directly and indirectly stimulates marine ecosystems across the Arctic Ocean through physical changes such as increases in melting sea ice and the open-water periods. A warming climate is also increasing access of the Arctic to industry, which is, in part, influencing a rapidly growing population. In turn, the rising population will lead to a rise in the inflow of the anthropogenic nitrogen compounds into the Arctic coastal areas. Indeed, part of the problem is logistical in nature, where wetland lagoon wastewater treatment systems that are relatively efficient in temperate zones have limited efficiencies due to low temperatures and polar darkness. Although there was no evidence of eutrophication in the system, due to the gradually increasing population of the region and associated wastewater effluent, there is a need to consider improvements to the wastewater treatment system. First, installation of secondary treatment should be considered. In particular, secondary treatment using seaweed or plants is environmentally friendly and efficient in removing nutrients. Some Arctic communities are attempting to develop secondary treatment in addition to the primary one (Ragush et al., 2015). Second, increasing the retention time of wastewater within the wastewater treatment system is one way of improving the treatment process. In particular, increasing retention time in lagoons can lead to more extensive microbial activity. Or, by increasing the size of the wetland, the absorption efficiency for the high nitrogen compounds contained in the wastewater by the wetland bottom and surrounding soil increases. Lastly, it is possible to think of ways to increase the period of wastewater discharge and to reduce the flow rate of discharge with increases in the open water period. If the amount of wastewater that inflows per second decreases, the more active ocean dilution can be achieved in the receiving water, ultimately resulting in less phytoplankton stimulation.

Through my case study in Cambridge Bay, I found that there are impacts of the wastewater effluent on phytoplankton biomass, species composition and local primary production. These findings highlight a need for future research and monitoring of the region and other similar Canadian coastal Arctic regions. Below I list some ideas of what can be done:

First, one of the most important findings was the important role of the persistent halocline and bathymetry that acted to trap nitrogen at depth in the bay and ultimately accounted for a large part of the bloom above background production rates. However, interannual evidence suggested this nitrogen inventory could periodically ventilate. The process behind this ventilation requires additional investigation. It is suggested to install small oceanographic moorings with current meters and CTDs on either side of the bay within the 20 m sill region to monitor advection rates and salinities of waters coming in and leaving the bay. An additional mooring can be placed in the centre of the bay to monitor the deep water salinities of the bay. It would also be fruitful to add biologically-relevant sensors, such as PAR sensors, *in vivio* Chl *a* fluorometers, and dissolved oxygen sensors, to these mooring to provide background information on primary production. Linked to the existing Cambridge Bay oceanographic observatory run by Oceans Network Canada, as well as a new deep water mooring in the bay, these data will be invaluable to investigate ventilation of waters in the bay.

Second, during the period of wastewater discharge, high concentrations of nutrients were not efficiently removed, and almost all of them were released into the bay. The influx of anthropogenic nutrients can cause eutrophication. Some previous studies focused on legacy nutrients, especially that of phosphorus, because anthropogenic phosphorus accumulates over long periods of time in receiving waters of freshwater systems (Carpenter, 2008; Sharpley et al., 2013). The result of legacy nutrients can be observed as mixing occurs in the system resulting in

intense surface blooms without the input of wastewater (Paerl, 2014; Paerl, et al., 2018). These types of systems can be compared quite closely to the persistent pycnocline and shallow exit sills of Cambridge Bay and thus the nitrogen accumulating at depth could cause a legacy nutrient condition if ventilation does not occur for longer periods of time. An additional concern is eutrophication of the system overtime, where large masses of settling organic matter and associated remineralization processes at depth can lead to oxygen depletion (Herbert, 1999; Voss et al., 2013). Therefore, additional research is needed to investigate potential roles of legacy nutrients and changes in oxygen levels in the bay. Firstly, collecting sediment core samples at the outfall and CB station, and then analyzing nutrients in sediments to make a time-series database to trace the amount of nutrients which accumulated from the wastewater influx over the years. After establishing the time-series database based on the core samples, the research for additional nutrient input and for oxygen concentration needs to be conducted each year to investigate whether the seawater is changing in ocean conditions in order to establish a monitoring program. This study, when conducted concurrently with the first study recommended above, will help us better understand the ocean conditions affected by human activities as well as climate-induced change in the region.

Third, similar research to that accomplished in this thesis should be periodically repeated year by year in order to establish a monitoring program related to the interaction between regional primary production and the amount of annual wastewater effluent. Such a monitoring program could also be conducted alongside environmental assessment focusing on wastewater contaminants such as pharmaceuticals, antibiotics resistance genes, and microplastics. Furthermore, even though phytoplankton species such as *Psuedo-nitzschia spp.*, which can cause harmful algal blooms (Pan and Rao, 1997), were not observed in 2018, their occurrence is a

possibility as they are a cold-water species and have been reported elsewhere in the Arctic (AMAP 2017). Establishing a monitoring system will enable observations of changes and trends in the system, which will undoubtedly help local decision-makers manage the wastewater treatment system better.

Last, more case studies like this thesis research should be conducted across the coastal Arctic. Not all coastal communities are comparable to Cambridge Bay. In fact, the semi-enclosed fjordal-type bay is likely unique among Canadian coastal communities. However, many other communities are located in protected embayments and could be influenced by local wastewater effluent. Furthermore, population size, wastewater treatment and collection systems in each community can differ greatly in their type and usage. A broader understanding on the impact of wastewater effluent on the coastal Arctic marine ecosystem will help improve our understanding for informed decisions to improve wastewater treatment systems and strategies.

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APPENDIX A: ABBREVIATIONS

Adenosine diphosphate	ADP
Adenosine triphosphate	ATP
Ammonium	NH_4^+
Analysis of variance	ANOVA
Cambridge Bay	CB
Chlorophyll *a*	Chl *a*
Chlorophyll *b*	Chl *b*
Conductivity, Temperature, Depth	CTD
Deep chlorophyll maximum	DCM
Filtered seawater	FSW
High performance liquid chromatography	HPLC
Nitrate	NO_3^-
Nitrite	NO_2^-
Nitrogen	N
Organic matter	OM
Particulate organic carbon	POC
Particulate organic matter	POM
Particulate organic nitrogen	PON
Phosphate	PO_4^{3-}
Phosphorous	P
Photosynthetically active radiation	PAR
Queen Maud Gulf	QM
Silicic acid	$Si(OH)_4$
The west of Dease Strait	WD
The east of Dease Strait	ED
Total nitrogen	TN