

CHAPTER I

INTRODUCTION

Background and Significance

Cougar Lake on the campus of Southern Illinois University Edwardsville has received both thermal effluent from the campus' heating/refrigeration plant and treated wastewater from the treatment facility for the entire campus since its construction in 1965. Although wastewater at the facility undergoes primary, secondary, and tertiary treatment, nutrients such as nitrogen have not always been removed during those processes (Brady 1999, Deming 2000, Rieger 2003). Because it is not regulated by the IEPA, nitrogen remains in the wastewater after treatment and is discharged into Cougar Lake. Excessive nutrients and high organic production in Cougar Lake cause it to be classified as a eutrophic lake, meaning primary productivity is extremely high because nutrient levels are also high (Wetzel 1983). Eutrophic lakes also display depleted oxygen levels in the cool water at the bottom of the lake, the hypolimnion, after only a couple of weeks of stratification, resulting in continuous anaerobic conditions throughout the summer (Wetzel 1983). Eutrophication is becoming more widespread in lakes because of the impact of anthropogenic activities, such as pollution in the form of runoff from sewage and fertilization. The shift from low to high primary productivity in lakes is usually caused by excess nitrogen and phosphorus (Wetzel 1983).

Seasonal temperature also influences the onset of stratification, which is the distinct layering of the water in lakes typically lasting from spring until the fall. Increased surface water temperature in the spring creates a thermocline where the less dense warm water floats on top of the cool dense water of the hypolimnion. Since the hypolimnion is cut off from the well-oxygenated epilimnion, or top layer, its temperature remains cool throughout the summer months until the fall turnover when it again mixes with the rest of the lake. As the

cooler dense water is confined to the bottom of the lake, it quickly becomes oxygen-depleted and all physiological processes become anaerobic.

Water from Cougar Lake is used in the campus heating/refrigeration plant, and when it is discharged back into the lake the temperature is potentially different than it was before. Thermal effluents can also cause stratification for extended periods of time, which also affects the physical mixing of nutrients and waters in the lake. Stratification typically starts to appear in the spring and lasts until cooler weather approaches in the fall, however the input of thermally treated water into the lake can extend the period of stratification.

Natural abundance studies of stable isotopes and their distribution in aquatic ecosystems can indicate nutrient cycling or sources of carbon and nitrogen utilized by organisms (Goericke *et al.* 1994). Natural abundance studies utilizing stable carbon and nitrogen isotopes of various materials such as sediments, seston, and particulate organic matter in lakes, estuaries, and oceans have given indications of both source material and physical processes (Vuorio *et al.* 2002, Graham *et al.* 2001, Bernasconi *et al.* 1997, Thornton and McManus 1994). The analysis of carbon isotopes can give indication of the type of photosynthetic pathway and the carbon source of the material while analysis of nitrogen isotopes can reveal sources of inorganic nitrogen, such as inorganic fertilizers or animal waste.

Hypothesis

This study was conducted to characterize the annual cycling and changing isotopic composition of seston in Cougar Lake using carbon and nitrogen stable isotopes. Influences such as temperature and oxygen will be discussed, as well as the impact of both the

wastewater treatment facility and heating/refrigeration plant on campus. Questions addressed in this study are:

1. Can changes in the isotopic composition of seston be attributed to seasonal changes in temperature and dissolved oxygen that occur on a regular basis?
2. Does the effluent from the campus-based wastewater treatment facility influence nitrogen isotope values throughout the year or only during times of high primary productivity?
3. Is there any indication from the heating/refrigeration plant on the isotopic composition of seston other than its possible influence on lengthening the stratification period?
4. Do carbon isotope values provide any information other than the source of the material?

Based on previous background information, it is hypothesized that the nitrogen isotopic composition of the wastewater released into Cougar Lake will impact the isotopic composition of seston in the lake throughout the year, especially during periods of high primary productivity and stratification. The impact should be revealed throughout the period of stratification, causing increased $\delta^{15}\text{N}$ values. Carbon isotopes are also expected to change with productivity levels, which can be related to seasonal changes such as temperature and oxygen levels.

To identify the patterns of stratification, photosynthesis, and source materials in the water column of Cougar Lake and the processes that control carbon and nitrogen isotopic abundances, seston was collected at meter-depth intervals starting at the surface and continuing to the bottom of the lake (11 meters) at monthly intervals for a period of one

calendar year. Seston was separated from the water through filtration and analyzed for $\delta^{15}\text{N}$, %N, $\delta^{13}\text{C}$, and %C at each depth-interval and compared to corresponding temperature and dissolved oxygen readings from the lake.

CHAPTER II

STUDY AREA

Cougar Lake was constructed in 1965 on the campus of Southern Illinois University Edwardsville to provide cooling water for the University's heating/refrigeration plant (Lajeone 1972). The lake's surface area is 31.4 ha, its average depth is 3.9 meters, and its watershed area is approximately 242.9 ha, the majority composed of grassland (Lajeone 1972). The water intake for the heating/refrigeration plant is approximately 45.7 meters from the shore and the discharge flume is 804 meters, 1.5 meters wide, 1.8 meters deep, and drops 7.6 meters from start to finish (Lajeone 1972). Discharge from the heating/refrigeration plant back into the lake is north of the intake area.

Cougar Lake also receives effluent waters from the University's wastewater treatment facility near the dam on the north side of the lake. In addition to primary and secondary treatment, the effluent undergoes a tertiary treatment that removes phosphate via precipitation with alum. Nitrogen is not removed from the effluent, and high concentrations of nitrate remaining in the effluent can contribute to eutrophication of surface waters (Macko and Ostrom 1994). Previously, copper sulfate treatments have been used to control algal blooms in Cougar Lake, as much as 17-22.5 kg per surface hectare three times in 1971 (Lajeone 1972) and approximately 9 kg per surface hectare twice in 1975 (Rosen 1978). More recently, between 1990 and 1999, 348 kg of copper sulfate was applied per year (Guo 2002). Application was suspended in 2000 (Guo 2002). Based on the recommended application rate of 6.07 kg/ha for high alkalinity waters, Cougar Lake was treated with twice the recommended amount of copper sulfate for over two decades (Guo 2002). Currently, copper sulfate is not used for algal control.

Cougar Lake is a typical eutrophic lake, which describes a lake that has an increased supply of nutrients and high productivity (Wetzel 1983). Rising temperatures in the spring induce stratification where the temperature gradient between shallow and deep waters increases and contains all aerobic processes in shallow waters. The epilimnion is the shallow warm water layer that is heated by solar radiation. During times of high productivity, solar radiation can only warm the water to a certain depth, which creates a thermocline where water temperature decreases exponentially with increasing depth. The cooler, denser water at the bottom of the lake is termed the hypolimnion. During this stratification period, wind currents circulate the upper layer of warmer water. In the fall the water column destratifies and the deeper waters are then circulated to the shallow waters. In his study of Cougar Lake, then called Tower Lake, Lajeone (1972) first documented thermal stratification on March 13, 1972, which held until September 20th, and the fall turnover was complete by November 8th. Cougar Lake undergoes one period of complete mixing, typically between November and March (Reiger 2000).

Oxygen levels follow a similar stratification period in a eutrophic lake such as Cougar Lake. In the epilimnion, the photosynthetic production of organic matter and water circulation via wind currents replenishes the oxygen levels during stratification. The hypolimnion is severely depleted of oxygen, mainly from the biological oxidation of organic matter, especially at the water-sediment interface, and from plant and animal respiration (Wetzel 1983). The depletion of oxygen in the hypolimnion gives rise to anaerobic processes by bacteria and drastically reduces the habitable area of plants and animals the lake. As the fall turnover begins, the well-oxygenated waters are circulated deeper into the hypolimnion and oxygen levels once again increase to levels limited only by temperature.

The discharged effluent from the heating/refrigeration plant has also influenced temperature in Cougar Lake. Discharge volume and temperature from the plant is seasonably variable as most water intake occurs in the summer months when cooling demand is high, and subsequently this variation impacts the temperature of Cougar Lake. Effluent from the heating/refrigeration plant is greatly increased in volume and temperature between April and September, resulting in an extended thermal stratification period (Lajeone 1972).

Previous limnological studies (Lajeone 1972, Rosen 1978) and studies involving the cycling of minerals at Cougar Lake (Brady 1999, Deming 2000) can be used as guides for the study of seston. Brady (1999) and Deming (2000) both concluded that elements such as phosphorus, silica, iron, manganese, and sulfide are recycled into the water column from the bottom sediments of Cougar Lake. Brady (1999) also concluded that the recycled nutrients from the bottom of the lake make a significant contribution to the high trophic status of the lake. It is hypothesized that nitrogen and carbon isotopic composition of seston can be influenced by the cycling of these minerals from the bottom of the lake back into the water column.

CHAPTER III

REVIEW OF RELATED LITERATURE

Nitrogen in Aquatic Ecosystems

Nitrogen is an important nutrient and natural contaminant in aquatic ecosystems such as Cougar Lake. It is present in forms such as nitrate (NO_3^-), nitrite (NO_2^-), ammonium (NH_4^+), ammonia (NH_3), and nitrogen gas (N_2) and can be included many transformations, including nitrogen fixation, mineralization, volatilization, nitrification, and denitrification. The mineralization of organic nitrogen into ammonium can lead to both volatilization of nitrogen in the atmosphere and the oxidation to nitrite and nitrate (Macko and Ostrom 1994). Nitrate is an important pollutant in groundwater especially as human ingestion of nitrate-contaminated water can lead to a medical condition called methaemoglobinaemia, or “blue baby syndrome” which deprives blood of its oxygen (Macko and Ostrom 1994). Nitrate sources for both groundwater and surface waters include fertilizer nitrogen, natural soil-derived nitrate, and animal waste/sewage-derived nitrate (Macko and Ostrom 1994). With increased inorganic fertilizer use and human population growth, nitrate contamination can be a significant threat to all waters, including groundwater and surface water.

The amount and types of nitrogen inputs in an aquatic ecosystem, such as Cougar Lake, vary throughout the season. Migratory fowl in densities as much as one per m^2 that are present for a brief period can contribute significant amounts of nitrogen and phosphorus from the guano left behind (Wetzel 1983). The presence of geese on the SIUE’s campus is therefore a potential contributor of guano nitrogen and phosphorus into the lake. Other seasonal contributors of nitrogen are fertilizer runoff from adjacent agricultural lands and sewage input from the campus. Fertilizers for both agricultural and landscaping purposes are

generally applied in the spring, adding to the nutrient loading of the lake at that time of the year. The effluent volume from the sewage treatment facility on campus also fluctuates (Table 1) over the course of the year. The sharp increase in effluent discharged per day between August and September is concurrent with the increase in student population on campus because many student residents are absent in the summer months. The decreased effluent in December also corresponds to the decrease in student population between the fall and spring semester. The amount of treated wastewater discharged into Cougar Lake impacts the amount of nitrogen being released into the lake and it may also influence the nitrogen isotopic composition of seston. The forms, sources, and seasonal variations of nitrogen are important to this study because each has an important impact on the isotopic composition of seston.

Table 1: SIUE Wastewater Treatment Facility Effluent Averages.

Date	Effluent Avg. (L/day)	NH₃ Avg. (mg/L)	Effluent Avg. (L/month)	NH₃ Avg. (kg/month)
Mar-03	840,000	1.20	26,000,000	31
Apr-03	1,100,000	4.99	32,000,000	160
May-03	720,000	0.16	22,000,000	3.6
Jun-03	700,000	0.19	21,000,000	4.0
Jul-03	670,000	0.10	21,000,000	2.1
Aug-03	770,000	1.30	24,000,000	31
Sep-03	1,200,000	0.91	36,000,000	33
Oct-03	1,000,000	0.63	34,000,000	21
Nov-03	890,000	0.81	27,000,000	22
Dec-03	690,000	0.87	21,000,000	19
Jan-04	750,000	0.67	23,000,000	16
Feb-04	990,000	0.26	29,000,000	7.5
Mar-04	1,000,000	0.20	31,000,000	6.2

Seston

The purpose of this study is to determine the origin of nitrogen and carbon in seston in the water column at Cougar Lake, its isotopic composition, and seasonal fluctuations with the combined use of carbon and nitrogen stable isotopes. Seston is all suspended material, both organic and inorganic particles, able to be collected on a filter and primarily consists of small particles with water column residence times on the order of several hundred years (McCusker *et al.* 1999, Huang *et al.* 2003). Seston consists of mineral grains, phytoplankton cells and fragments, microzooplankton, amorphous inorganic and organic matter, coal fragments, bacteria, small fecal pellets and fibers (McCusker *et al.* 1999). Cole *et al.* (2004) found that nonvascular macroalgal $\delta^{15}\text{N}$ values were a better predictor of wastewater than vascular macrophytes because they more closely reflected increases in water-column dissolved inorganic nitrogen (DIN) values. Because seston remains in the water column for a very long time, stable isotope analysis can be a useful tool in tracking its origin in the water column at Cougar Lake from season to season.

Stable Isotopes and Analysis

Stable isotopes are atoms of an element with the same number of protons and electrons, but with different numbers of neutrons, thereby affecting the mass and bond energy. Unlike radioactive isotopes, stable isotopes do not decay. Stable isotopes that have low atomic numbers are used in light stable isotope geochemistry because the relative mass difference between the rare (heavy) and abundant (light) isotope is large, and these differences can cause an element to have slightly different chemical and physical properties (Kendall and Caldwell 1998). Carbon and nitrogen fall into this category as do hydrogen,

oxygen, and sulfur. Mass differences between isotopes are important because physical, chemical, and biological processes or reactions can cause fractionation, which is the change in the relative proportions of an isotope of the sample element in various compounds (Kendall and Caldwell 1998). Kinetic isotope effects such as evaporation and bacterial interaction are rapid and non-reversible and cause very large fractionations, whereas equilibrium reactions are reversible and fractionations are not quite as significant (Kendall and Caldwell 1998). Biological reactions involving organisms tend to utilize the lighter isotopic element because its bond energy is lower and more easily broken than heavy isotopes which lead to high fractionations, especially when the reaction pathway is slow and has more time to be selective or when the element utilized in the reaction is limiting.

Several materials in the environment are known to have a specific isotopic “signature” based on the parent material or the physiological processes that generate the signature (See Table 2). Different isotopic signatures of nitrogen can occur under certain circumstances. The most distinct nitrogen isotope signature comes from wastewater or animal manure, which has a $\delta^{15}\text{N}$ value of nitrate in the range of +10 to +25 ‰ (Kendall 1998). The $\delta^{15}\text{N}$ values are higher because physiological processes involved with the breakdown and assimilation of food in animals (including humans) tend to utilize the lighter isotope (^{14}N), leaving behind an increased amount of ^{15}N in the manure or wastewater. The $\delta^{15}\text{N}$ of nitrate in groundwater ranges from +2 to +8‰ (McClelland and Valiela 1998).

The carbon isotopic “signature” can also give indication of parent material in terrestrial ecosystems or sources of carbon from dissolved organic carbon in groundwater. Most terrestrial plants, which includes all trees, most shrubs and cool-season grasses, utilize the C_3 photosynthetic pathway and tissues from these plants have a $\delta^{13}\text{C}$ value ranging from

–20‰ to –35‰ (Wang *et al.* 1998). Another category of plants, which include warm-season grasses and a few shrubs, utilize C₄ photosynthesis and tissues from these plants contain δ¹³C values of –9‰ to –17‰ (Wang *et al.* 1998). The δ¹³C values of dissolved organic carbon in groundwater tend to reflect the average δ¹³C of local dead or decomposing plant material in the area, between –26‰ and –30‰ where C₃ plants are predominant and have values up to –18‰ where C₄ plants dominate (Wang *et al.* 1998).

Table 2: Nitrogen and Carbon Stable Isotope Values of Various Source Materials.

<u>Source</u>	<u>δ¹⁵N</u>	<u>δ¹³C</u>	<u>Reference</u>
Soil	-10 to +15		Kendall 1998
Plants	-5 to +2		Kendall 1998
Groundwater derived from fertilizer	-3 to +3		Cole <i>et al.</i> 2004
Atmospheric N	0 to +3		Kendall 1998
Groundwater from natural soils	+2 to +8		McClelland <i>et al.</i> 1998
Groundwater derived from atmospheric N	+2 to +8		Cole <i>et al.</i> 2004
Wastewater	+10 to +20		McClelland <i>et al.</i> 1998
Wastewater	+10 to +22		Cole <i>et al.</i> 2004
Barnyard soil	+10 to +22		Macko & Ostrom 1994
Animal Manure	+10 to +25		Kendall 1998
C ₃ plants		-20 to -35	Wang <i>et al.</i> 1998
CAM plants		between C ₃ and C ₄ values	Wang <i>et al.</i> 1998
C ₄ plants		-9 to -17	Wang <i>et al.</i> 1998
Atmospheric CO ₂		-8	Wang <i>et al.</i> 1998

Analysis of stable isotopes is distinctly different from elemental analysis because the *ratios* of two isotopes of the same element, typically the isotopically heavy isotope versus the light isotope (usually the most abundant), are compared to a reference standard. The material

in question is analyzed simultaneously with the reference standard instead of consecutively. These results are expressed as delta (δ) and are designated in “per mil” (‰) values determined by the following formula:

$$\delta_x = (R_x - R_{\text{std}} / R_{\text{std}}) \times 10^3;$$

Where R_x is the ratio of the heavy isotope to the light isotope of an element (e.g., $^{15}\text{N}/^{14}\text{N}$) and R_{std} is the same isotopic ratio of a reference standard. Atmospheric nitrogen is assigned a 0‰ value and is the reference standard for ^{15}N . Pee-Dee Belemnite (limestone) is assigned a 0‰ value and is the reference standard for ^{13}C . Results from this formula generate both positive and negative values that are distinctly different and very important for interpretation. A positive result means that the isotopic ratio of the sample is higher than that of the standard, whereas a negative result means the isotopic ratio of the sample is less than that of the reference standard.

Nitrogen Isotopes

Nitrogen in the water column is present in many forms, including nitrate (NO_3^-), nitrite (NO_2^-), ammonia (NH_3), ammonium (NH_4^+), and dissolved nitrogen gas (N_2). Processes that convert one form of nitrogen into another will cause changes in $\delta^{15}\text{N}$, either positively or negatively. Nitrogen isotope values of seston are influenced by the $\delta^{15}\text{N}$ values of different particles that contribute to its composition, the isotopic composition of the dissolved inorganic nitrogen source, and fractionation associated with transformation processes of one form of nitrogen to another (McCusker *et al.* 1999). $\delta^{15}\text{N}$ of organic matter increases approximately 3-4‰ with each trophic level transfer (Hodell and Schelske 1998).

Some transformation processes that cause enriched $\delta^{15}\text{N}$ values include assimilation of ^{15}N -enriched NH_4^+ , microbial degradation, microzooplankton grazing, peptide bond hydrolysis, formation of dissolved organic nitrogen, and the disaggregation of sinking particulate organic matter (McCusker *et al.* 1999). Processes that can lower $\delta^{15}\text{N}$ values of seston are assimilation of ^{15}N depleted NO_3^- , N_2 fixation, and disaggregation of sinking particulate organic matter (McCusker *et al.* 1999). The difficulty lies within the ability to identify which processes are influencing the fractionation of ^{15}N , which makes it very difficult to identify the source of the nitrogen.

The identification of processes that dictate $\delta^{15}\text{N}$ in lakes has been the focus of previous research. In a study of Lake Superior (Ostrom *et al.* 1998), seston was never found to have a lower $\delta^{15}\text{N}$ value than that of the nitrate present, which is the primary source of inorganic nitrogen available to phytoplankton. Nutrient assimilation by phytoplankton should result in $\delta^{15}\text{N}$ of seston less than or equal to that of the $\delta^{15}\text{N}$ of inorganic nitrogen (Ostrom *et al.* 1998). Therefore, if $\delta^{15}\text{N}$ of seston is higher than $\delta^{15}\text{N}$ of the nutrient source to the phytoplankton, another process is either solely or partially responsible for the $\delta^{15}\text{N}$ signature of seston.

Low $\delta^{15}\text{N}$ values of seston occur at depths in the water column where nitrate is not limiting (Ostrom *et al.* 1997). When nitrate is not the limiting factor at a particular depth in the water column, phytoplankton tends to uptake the lighter nitrogen isotope during assimilation, which results in lower $\delta^{15}\text{N}$ of seston. At this time seston will attain a lower isotope signature than the nitrate available in the water column. Low $\delta^{15}\text{N}$ values recorded at Lake Superior were found to reflect either an input of depleted ^{15}N from an outside source or fractionation during microbial processes that concentrate ^{14}N in nitrate (Ostrom *et al.* 1998).

Phytoplankton growing where nitrate is limiting do not discriminate between the light and heavy isotopes, and therefore assume a similar $\delta^{15}\text{N}$ to nitrate (Ostrom *et al.* 1997).

Depth within the water column is another variable that will determine the nitrogen isotope composition of seston. Depth influences the dissolved oxygen content of water in lakes, especially in the summer when thermal stratification restricts the movement of oxygen to the hypolimnion. The reduced dissolved oxygen content in the hypolimnion changes the microbial processes from aerobic to anaerobic, which in turn changes the $\delta^{15}\text{N}$ of seston. The vertical distribution of $\delta^{15}\text{N}$ of dissolved nitrate in Lake Lugano, Switzerland, showed increased $\delta^{15}\text{N}$ as nitrate concentration decreased in the hypolimnion, which suggested microbial nitrate reduction accounts for the nitrate removal (Lehman *et al.* 2003). Lehman *et al.* (2003) assumed that since the hypolimnion is a closed system during the presence of a thermocline, processes such as nitrate assimilation and external nitrate loading occurring in the epilimnion do not affect the nitrate levels in the hypolimnion. Seasonally, primary production has an effect on nitrogen isotopes in the water column. At Conception Bay, Newfoundland, seston with a high $\delta^{15}\text{N}$ were more common in the spring than at other times, and at a depth just below the chlorophyll fluorescence maximum (Ostrom *et al.* 1997). Water stratification occurring seasonally will also affect nitrogen isotopic composition in the water column. In the presence of a well-defined thermocline, the epilimnion remains well-mixed by surface winds, essentially creating a closed system in the hypolimnion.

Input of wastewater into lakes and other water bodies also has an effect on $\delta^{15}\text{N}$. Both volatilization of ammonia and denitrification remove ^{14}N at a faster rate than ^{15}N , resulting in $\delta^{15}\text{N}$ of nitrate in the wastewater entering the water body between +10 and +20‰ (McClelland and Valiela 1998). Indirect inputs of wastewater can also affect the $\delta^{15}\text{N}$ of

adjacent water bodies, as discovered by Griggs *et al.* (2003). In the Florida Keys, treated wastewater is directly injected into the groundwater through six gravity-driven wells at the rate of 500-1200 m³ day⁻¹. The highest nitrate concentration of groundwater was found closest to the point of injection with the exception of one sampling location that was the furthest east (Griggs *et al.* 2003). This zone of elevated nitrate concentrations occurred at 9 meters, with lower nitrate concentrations occurring both above and below this zone. The $\delta^{15}\text{N}$ values of wastewater in four samples ranged from +7 to +20‰ and $\delta^{15}\text{N}$ values in the fresh and brackish waters (as determined by salinity) ranged from +16 to +26‰ (Griggs *et al.* 2003). These high values in the fresh and brackish waters were consistent with ¹⁵N enrichment caused by partial denitrification, which, along with dilution, was considered to be the only processes affecting nitrate in groundwater. Because of this, the denitrified groundwater samples were determined to have an initial $\delta^{15}\text{N}$ values between +6 and 17‰, which was similar to the range measured in the wastewater samples.

Carbon Isotopes

Carbon isotopes are important in determining the sources of CO₂ utilized by phytoplankton and changes in levels of primary productivity (McCusker *et al.* 1999). These changes can be observed seasonally and at various depths in the water column of water bodies. $\delta^{13}\text{C}$ of plants can provide information on the sources of nutrients and food web relationships, especially when combined with other isotopes, such as $\delta^{15}\text{N}$ and/or $\delta^{34}\text{S}$ (Kendall 2004). Variable ¹³C values of seston can be attributed to factors such as different sources of seston, isotopic fractionation during carbon fixation, different phytoplankton species, pH, temperature, and dissolved inorganic carbon isotopic composition (McCusker *et*

al. 1999). Phytoplankton can change the carbon isotopic composition of seston by discrimination against ^{13}C in photosynthesis during diffusion of CO_2 into the cell (McCusker *et al.* 1999). Carbon that is fixed in plant tissue during photosynthesis is significantly depleted in ^{13}C relative to the atmosphere because plants discriminate against the heavier carbon isotope in the process of fixing carbon from atmospheric CO_2 into organic matter (Wang *et al.* 1998). Areas with enriched ^{13}C can be related to high rates of primary productivity if high chlorophyll and low dissolved inorganic carbon concentrations support that conclusion (Ostrom *et al.* 1997).

Carbon isotopic composition of seston is seasonally variable. The main factors controlling productivity are temperature and light, which are both significantly decreased in lakes during the winter months. Dissolved CO_2 is the main source of C for most algae, and seasonal changes in temperature and temperature-dependent fractionation between bicarbonate and dissolved CO_2 will induce a change in isotopic composition of carbon (Bernasconi *et al.* 1997). Seston at Conception Bay, Newfoundland, contained high $\delta^{13}\text{C}$ values that are associated with low dissolved inorganic carbon concentrations, and are most commonly linked to the spring algal bloom (Ostrom *et al.* 1997).

Similarly, in a study of Lake Ontario, Hodell and Schelske (1998) found that the $\delta^{13}\text{C}$ of bulk organic matter in the water column increased after the onset of stratification because of high rates of production and draw down of $^{12}\text{CO}_2$ from the epilimnion. Prior to thermal stratification, carbon isotopic values of phytoplankton were low during the spring months (March – May) because nutrients are utilized throughout the water column.

Depth within the water column is also a controlling factor of carbon isotopic composition. As a component of seston, organic matter's carbon isotopic composition is

dependent on its source of dissolved inorganic carbon, which also varies with its response to the removal of ^{12}C during organic matter synthesis in the photic zone (Bernasconi *et al.* 1997). At Grand Traverse Bay, Lake Michigan, $\delta^{13}\text{C}$ increased from -28.4‰ at the surface to -26.2‰ at 20 M, which is also associated to similar trends of particulate organic carbon, particulate organic nitrogen, and NH_4^+ concentrations (McCusker *et al.* 1999).

Previous Studies with Carbon and Nitrogen Isotopes

Previous studies utilizing both carbon and nitrogen stable isotopes have focused on the analysis of sinking organic matter and sediments in lakes. Bernasconi *et al.* (1997) studied the annual cycle and seasonal carbon and nitrogen isotopic composition of sediment in Lake Lugano, Switzerland. The study involved the use of sediment traps at two depths, and the results of both traps revealed two distinct groups of organic matter: particles in the winter were characterized by heavy $\delta^{15}\text{N}$ (between $+10\text{‰}$ and $+16\text{‰}$) and very light $\delta^{13}\text{C}$ (between -31‰ and -40‰), and particles in the summer were characterized by light $\delta^{15}\text{N}$ (between $+3.6\text{‰}$ and $+8\text{‰}$) and light $\delta^{13}\text{C}$ values (between -22‰ and -30‰).

In the Forth Estuary, Scotland, Graham *et al.* (2001) characterized the isotopic composition of bottom sediments (between 0-10 cm) at different areas of the estuary in order to determine the source and the fate of incoming organic matter. Sediment samples were collected at various distances from the head of the estuary to provide isotopic evidence of both natural and anthropogenic inputs. Results of isotopic analysis presented very similar results over the entire length of the Forth Estuary. $\delta^{13}\text{C}$ ranged from -24.4‰ to -23.6‰ where the least negative values were obtained from the lower estuary sites, and $\delta^{15}\text{N}$ ranged from $+4.7\text{‰}$ to $+6.2\text{‰}$. Contrary to expected results at the Forth Estuary, $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of

sediment did not identify terrigenous sources of sedimented organic matter. The absence of increased sediment organic content and increased $\delta^{15}\text{N}$ near inputs such as sewage treatment facilities or industrial discharges indicates that efficient mixing processes are present in the Forth Estuary.

To portray the variability between both lakes and seasons, seston and plankton from two shallow lakes in Finland were analyzed in May and September 1999 for both carbon and nitrogen stable isotopes (Vuorio *et al.* 2002). In both lakes, $\delta^{15}\text{N}$ of seston was enriched in May compared to September, probably due to the dominance of N_2 -fixing cyanobacteria in September, which leads to $\delta^{15}\text{N}$ values of 0-2 per mil (Vuorio *et al.* 2002). Seasonal and between-lake differences in $\delta^{15}\text{N}$ suggest changes in either fractionation and/or cycling of nitrogen (Vuorio *et al.* 2002). The two lakes were dissimilar in $\delta^{13}\text{C}$ in the spring, but were within the same range in September. The similarities in low $\delta^{13}\text{C}$ of both lakes seemed to be controlled by phytoplankton processes when the population was large and water inflow to both lakes was low. Although the lakes were isotopically different in May, similarities were visible and easily explained in both $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in September because of the physiological processes that control carbon and nitrogen isotopic signatures of lakes.

Questions remain about the $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston from highly eutrophic lakes such as Cougar Lake. The combination of analysis of seston from each depth interval in Cougar Lake and a one-month sampling interval for a period of one calendar year should provide additional insight to the environmental factors that control the abundances of ^{15}N and ^{13}C contained in seston.

CHAPTER 4

METHODS AND MATERIALS

Water samples were collected from Cougar Lake at the north end of the lake near the dam, which is also near the discharge point from the wastewater treatment facility (Figure 1). The sampling location was selected because it is known to be the deepest part of the lake (Brady 1999). In this area lake depth varies around 12 meters for most of the year, depending on rainfall. Sampling occurred at or near 1-month intervals between March 2003 and March 2004, where the shortest interval between events was 21 days and the longest interval was 78 days due to ice on the lake early in 2004. Sampling was delayed from the December 2, 2003 sampling event until March 2, 2004, when the ice layer finally melted and

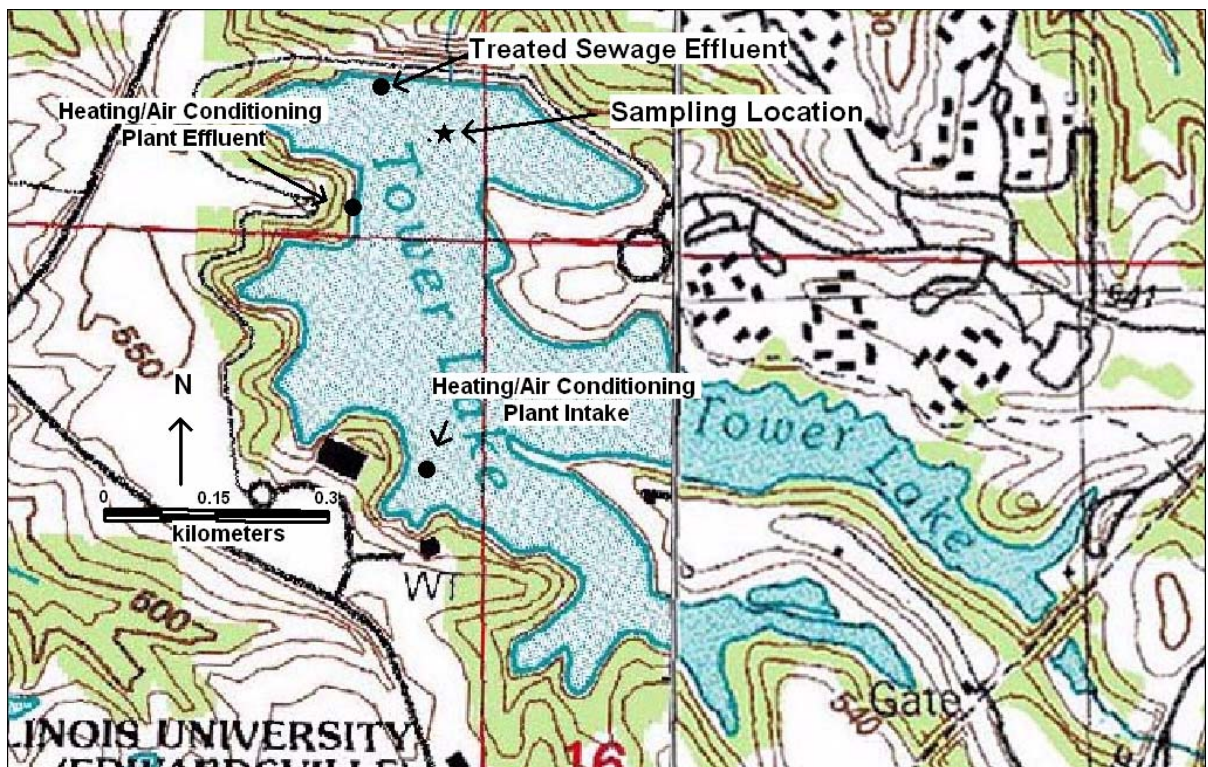


Figure 1: Cougar Lake.

a boat could again be used on the lake. A total of 13 sampling events were recorded during this study period.

Samples were collected with a 1.25 L polyethylene Kemmerer bottle at 1-meter depth intervals, beginning with the surface water (0 M) through the water column to 12 M unless water levels of the lake prevented the collection at 12 M. One liter of depth-interval sample was collected in acid washed Nalgene[®] bottles until seston could be separated in the lab. Temperature (°C) and dissolved oxygen (mg/L) readings were also recorded using a YSI oxygen probe at the same depth intervals, which provided information on the physical conditions of the lake at the time of sampling. Secchi disk readings were also recorded to indicate the level of light reaching into the water column.

On the same day of sampling, seston was isolated by filtering 400 mL of water through one Whatman[®] GF/C filter (47 mm diameter, 1.2 µm particle retention) under vacuum conditions. A total of two filters were used to separate seston from 800 mL of water at each sample depth. Both filters containing the seston were stored and dried in polyethylene centrifuge tubes at 80°C until they were dried completely and ready for freeze grinding. Samples were processed by grinding the combined filters from each depth interval in a Spex[®] Certiprep (6750 Freezer/Mill) Grinder. The freeze-grinding process was complete for each sample in approximately three minutes. After freeze grinding, each sample was transferred into a labeled 2.0 ml Micro Centrifuge tube. Upon completion all samples and a blank filter were sent to the Cornell-Boyce Thompson Stable Isotope Laboratory (CoBSIL) for isotope analysis of $\delta^{15}\text{N}$, %N, $\delta^{13}\text{C}$, and %C on the laboratory's ratio mass spectrometers.

Samples were analyzed at CoBSIL using a Finnigan MAT Delta Plus mass spectrometer that is interfaced through a Carlo Erba NC2500 elemental analyzer (EA) through a process that is known as continuous flow isotope ratio mass spectrometry. Carrier gas was Ultra high purity Helium (120 ml/min). Once the system attains vacuum, the instrument is tuned and background signals are checked for water, argon, N₂, and CO₂. Samples are then placed in an auto sampler on the EA. Samples are dropped into a combustion column that is packed with the oxidizing reagents, chromium and copper oxides. After combustion at 1000°C and a pulse of O₂ for complete and instantaneous combustion, the solid sample is converted to CO₂, N₂O, and H₂O. The N₂O is reduced to N₂ gas using copper wire in a reduction column set at 650°C. Water is removed using magnesium perchlorate in a water trap. After the water trap, remaining gases are run through a GC and then through an open split called a Conflo II. At the open split, the sample gases are directed into the mass spectrometer. This is where the reference gas injections of specific N₂ and CO₂ reference tanks are initiated. The entire process takes approximately 12 minutes per sample and provides data for % element and isotope ratio delta values.

CHAPTER V

RESULTS

Results are displayed in two types of graphs to visually illustrate the distribution of different variables in Cougar Lake. Vertical profiles give information of a single variable at one point during the study period and include different depths of the water column.

“Isopleths” resemble a contour map in that each line on the isopleth represents a single value of that variable throughout the depth profile and over time. Vertical profiles for each month of the study period, which includes temperature, dissolved oxygen, $\delta^{15}\text{N}$, and $\delta^{13}\text{C}$ data, can be found in Appendix B.

Temperature

Temperature (Figures 2 and 3, Appendix A) throughout the sampling period is an excellent indication of the physical limnology of Cougar Lake. Temperature readings taken on March 11, 2003 reflect the absence of stratification in Cougar Lake, which is typical throughout the winter months. The isothermal condition of the lake on this day showed temperatures ranging from 4.5°C to 4.7°C. The sampling event on April 11, 2003 was the first sign of stratification as the temperature at the surface of the lake rose from 4.7°C on March 11, 2003 to 18°C (see Appendix A) and the temperature at 11 meters rose only slightly from 4.5°C at the March sampling event to 8.4°C. The difference in temperature between the surface water (at 0 meters) and the bottom of the lake (at 11 meters) also increased between the months of March and April 2003. Whereas the temperature difference was only 0.2°C between the two depths of the lake in March, the difference increased in the span of one month to 9.1°C in April.

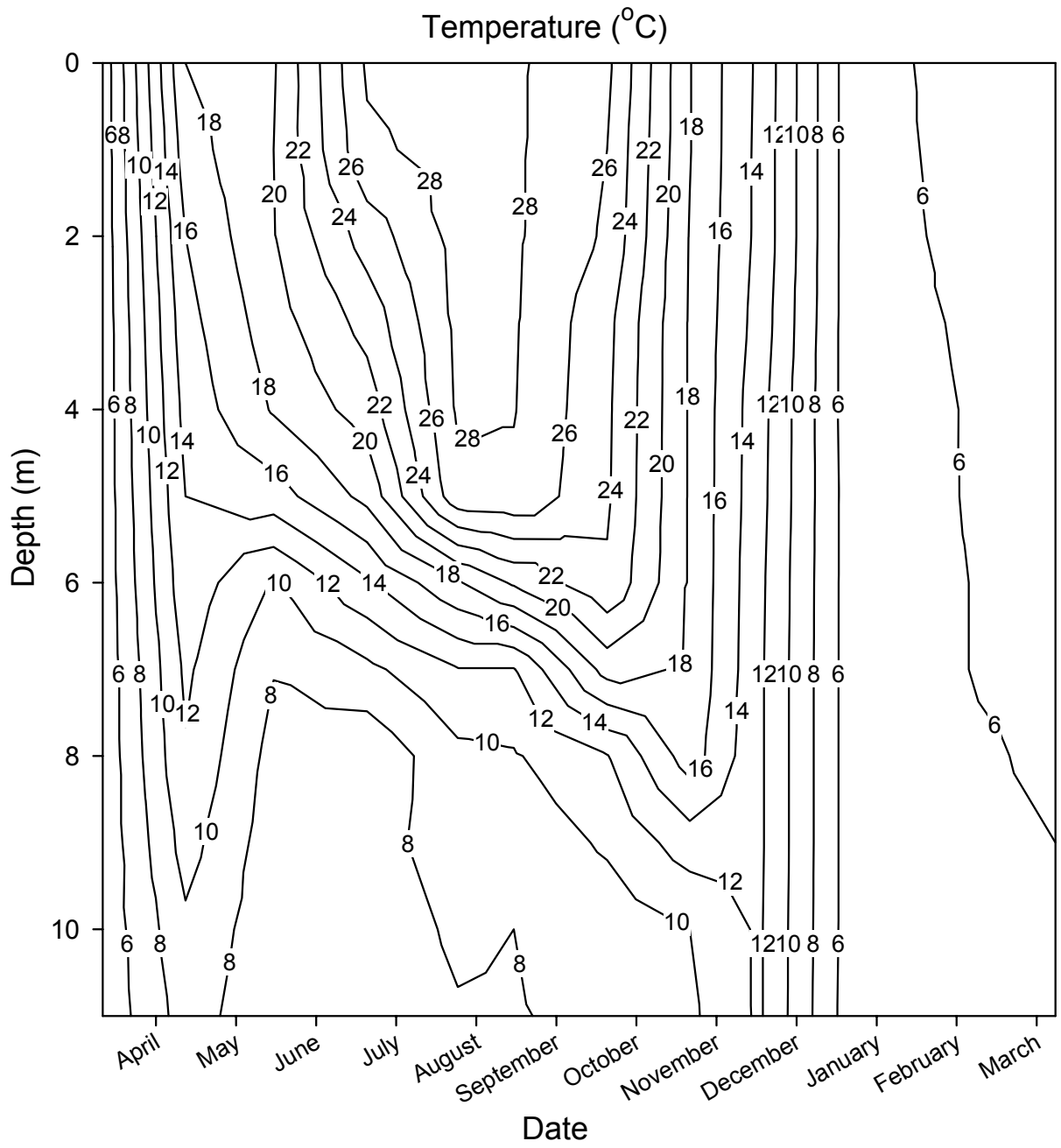


Figure 2: Temperature (°C) Contour Plot Diagram.

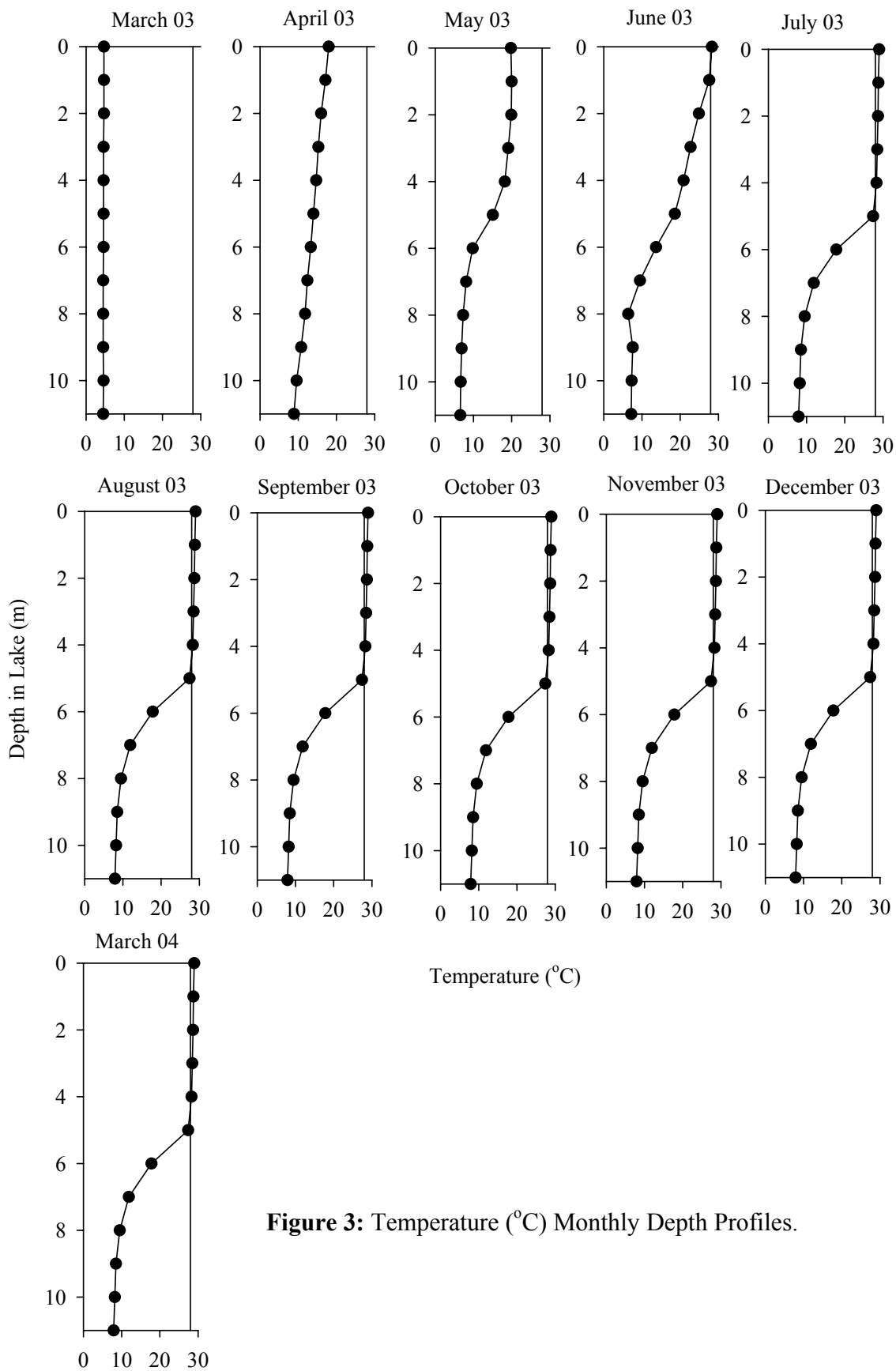


Figure 3: Temperature (°C) Monthly Depth Profiles.

Stratification of the lake was fully visible by the sampling event on May 18, 2003. Temperature ranged from 19.8°C at 0 meters to 15.1°C at 5 meters, decreased sharply to 9.8°C at 6 meters, and dropped slightly thereafter to 6.6°C at 11 meters. At this time the epilimnion was contained in the top five meters of the lake and remained through the summer months until the start of the fall turnover, which is visible in the October 17, 2003 sampling event. At this time, the temperature ranged from 18.1°C at the surface to 17.0°C at 8 meters. Temperature readings were noticeably more constant throughout the water column by the November sampling event, as the temperature difference between the surface and 11 meters was only 1.3°C. Isothermal conditions in the lake continued from November through the winter months and final sampling on March 2, 2004 when the temperature difference from the surface and 11 meters was only 2°C.

Dissolved Oxygen

Dissolved oxygen measurements showed the expected variation in concentration due to the changes in solubility with temperature (Figures 4 and 5, Appendix A). They also showed the depletion of oxygen due to heterotrophic metabolism in the hypolimnion. The dissolved oxygen readings indicate either aerobic or anaerobic processes at that particular depth in the lake, and stratification is noticeable from these readings on the isopleth diagram (Figure 4). Although the cool water of the hypolimnion is capable of holding more dissolved oxygen, it is cut off from the well-oxygenated water of the epilimnion during the summer and dissolved oxygen is quickly depleted. This initiates the shift from aerobic to anaerobic processes.

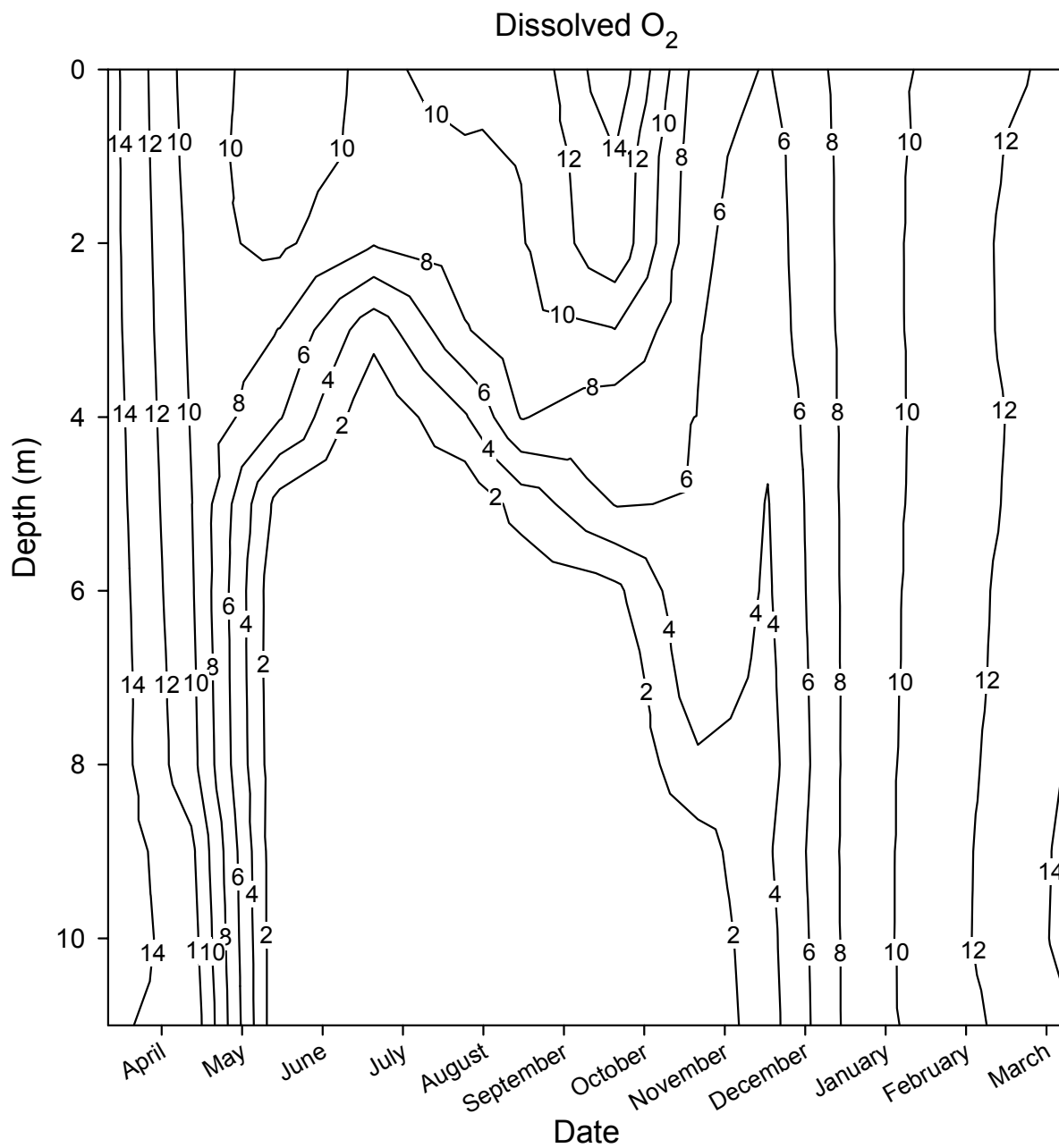


Figure 4: Dissolved Oxygen (mg/L) Contour Plot Diagram.

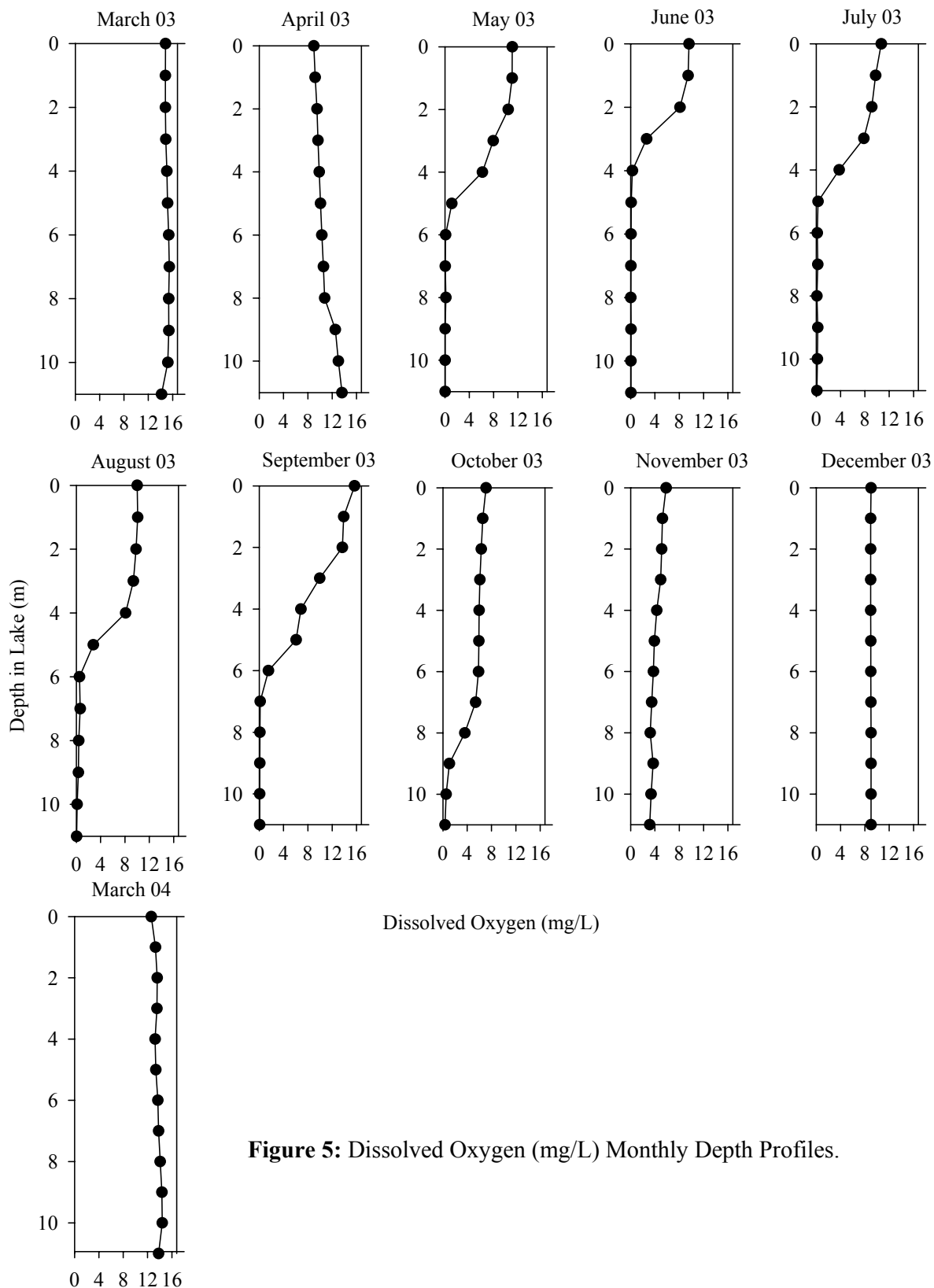


Figure 5: Dissolved Oxygen (mg/L) Monthly Depth Profiles.

At the beginning of the study in March 2003, dissolved oxygen ranged from 14.18 mg/L to 15.45 mg/L throughout the water column (Figures 4 and 5, Appendix A), and by the May 14, 2003 sampling event stratification was fully visible. At this time, dissolved oxygen levels of the surface (0 meters) was 11.05 mg/L, decreased to 1.13 mg/L at 5 meters, and remained below 1 mg/L at each depth from 6 meters to 11 meters. This pattern of anaerobic conditions below 5 meters in Cougar Lake continued as dissolved oxygen readings remained below 1 mg/L after the May sampling event and held through the summer months until August when it rose slightly to 2.8 mg/L.

Consistent with temperature readings in the lake, the fall turnover was first apparent on October 17, 2003 by the decrease in dissolved oxygen content at the surface waters from 15.67 mg/L in September to 7.11 mg/L in October. Dissolved oxygen readings were below 2 mg/L at 9, 10, and 11 meters in Cougar Lake in October, as opposed to the bottom six depth-intervals in the lake the previous five sampling events (May 14, 2003 through September 16, 2003). By November the difference in dissolved oxygen readings between the surface and the lowest depth-interval was 2.7 mg/L and in December the difference between the lowest and highest dissolved oxygen content of Cougar Lake was 0.07 mg/L, signifying the completion of the fall turnover process in Cougar Lake.

Nitrogen Isotopes

Nitrogen isotope values of seston in Cougar Lake (Figures 6 and 7, Appendix A) consistently revealed the physical characteristics throughout the seasons and depths of the water column. Nitrogen isotope values for the study period ranged from +3.75‰ (closest to

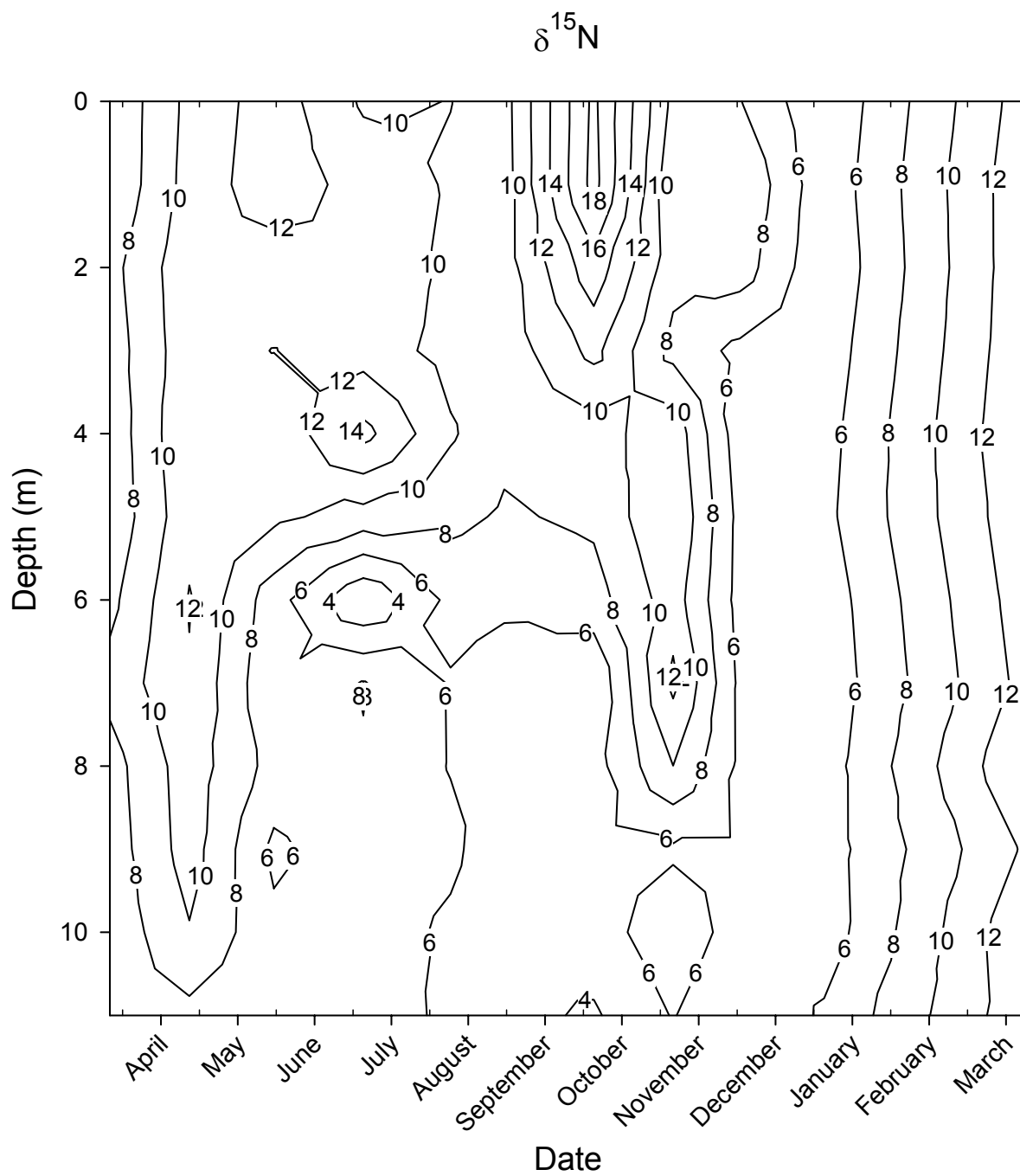


Figure 6: $\delta^{15}\text{N}$ Contour Plot Diagram.

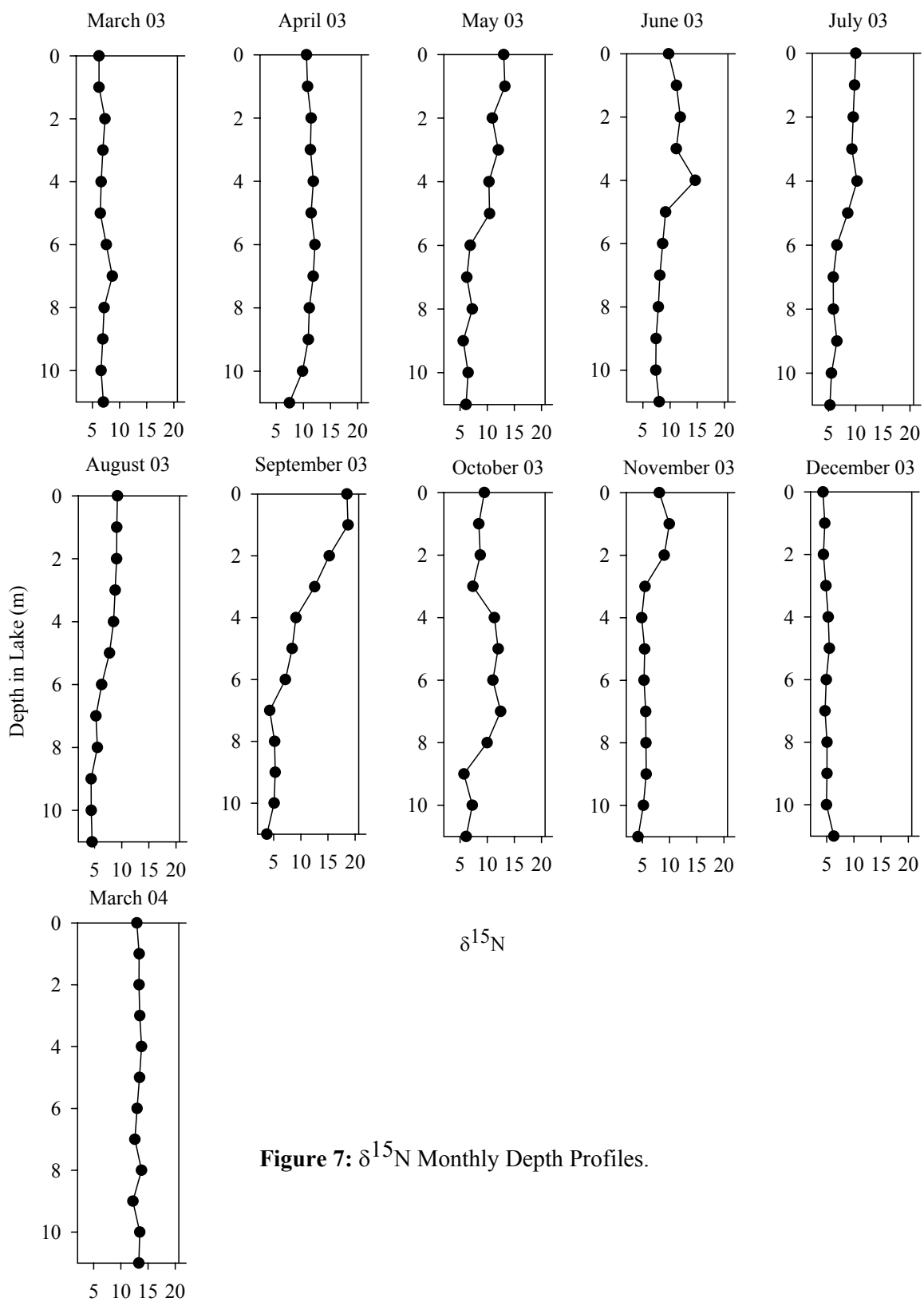


Figure 7: $\delta^{15}\text{N}$ Monthly Depth Profiles.

the signature of atmospheric nitrogen) at 11 meters to +18.7‰ (values consistent with those of animal waste) at 1 meter, both occurring on the September 16, 2003 sampling event. The blank filters analyzed with each sampling event contained no nitrogen with the exception of the blank sent with the August 2003 samples. At the first sampling event on March 11, 2003, $\delta^{15}\text{N}$ values were relatively constant throughout the water column. As time progressed from the first sampling event, the lake's surface water temperature rose with $\delta^{15}\text{N}$ values. For example, on the April 11, 2003 sampling event, $\delta^{15}\text{N}$ values rose throughout the water column from the previous sampling event, and these values also correspond to the increased temperature and high dissolved oxygen values throughout the water column. As the lake stratified in May 2003, the $\delta^{15}\text{N}$ values continued to rise in the top 5 meters of the lake where dissolved oxygen values remained above 1 mg/L. During this time, $\delta^{15}\text{N}$ signature of seston reflected that of animal waste. When nitrogen is not limited during photosynthesis, reactions involving primary producers preferentially uptake ^{14}N and leave behind the heavier nitrogen isotope, ^{15}N (Bernasconi *et al.* 1997). Bonds involving the lighter isotope require less energy to break, so while nitrogen is abundant the organism can choose between ^{14}N and ^{15}N . In this case, it will choose the lighter isotope because it requires less energy. When nitrogen is a limiting nutrient during photosynthesis, the organism generally will not discriminate between ^{14}N and ^{15}N .

This trend of high $\delta^{15}\text{N}$ values in the epilimnion continued into the fall until the October 17, 2003 sampling when the highest $\delta^{15}\text{N}$ values, which typically occurred at the surface of the lake throughout the summer, appeared at the lower part of the epilimnion (7-8 meters). At this depth, oxygen levels increased and temperature decreased slightly from the

previous month, revealing an expanded epilimnion since the sampling event in September 2003. In December 2003, $\delta^{15}\text{N}$ values decreased from the previous sampling event and returned to isothermal conditions, which ranged from 4.3‰ at the surface to 6.28‰ at 11 meters. At the March 2, 2004 sampling event, $\delta^{15}\text{N}$ values remained constant throughout the water column but increased to 12.93‰ at the surface to 13.29‰ at 11 meters. These high values previously occurred in the summer and early fall when the temperature was also higher.

Nitrogen isotope values followed similar patterns as temperature and dissolved oxygen readings throughout the water column and seasons. As the epilimnion was formed (already observed during the May 14, 2003 sampling event from the temperature data), oxygen was depleted in the hypolimnion and high nitrogen isotope values were contained in the epilimnion throughout the summer until the lake thoroughly mixed in November 2003.

Carbon Isotopes

Carbon isotopes of seston in Cougar Lake throughout the sampling period ranged from -20.55‰ at 10 meters in November 2003 to -31.97‰ at 2 meters in March 2004 (Figures 8 and 9, Appendix A) throughout the study period. However, blank filters analyzed with each month's samples contained carbon each month except for the September, October, and November blanks. The range of carbon isotopes indicates that seston originates from terrestrial plants (Table 2). Terrestrial plants with C_3 photosynthetic pathways have a $\delta^{13}\text{C}$ signature between -20‰ and -35‰ (Kendall and Caldwell 1998). Whereas temperature, dissolved oxygen, and $\delta^{15}\text{N}$ values recorded throughout the study period were indicative of

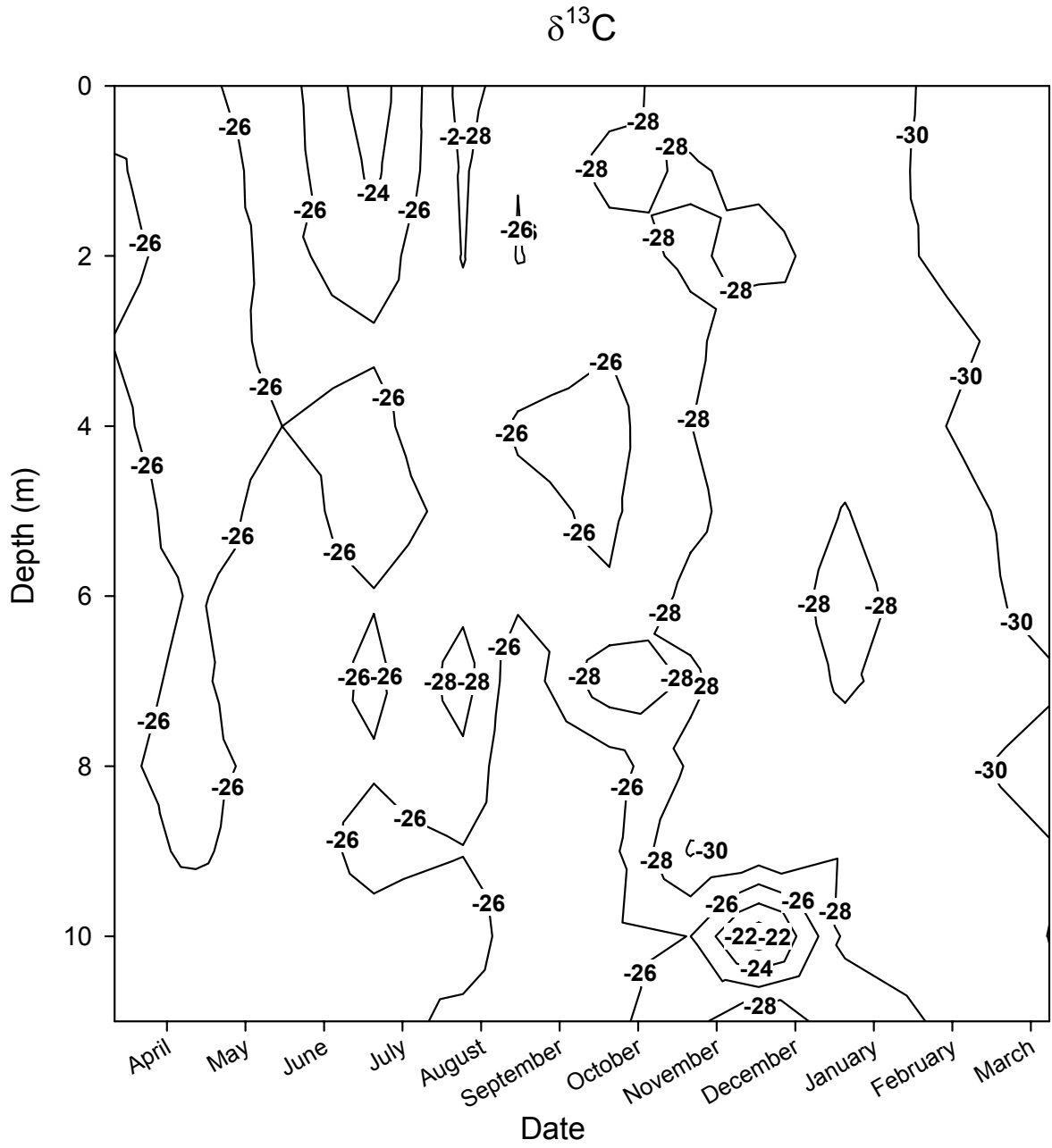


Figure 8: $\delta^{13}\text{C}$ Contour Plot Diagram.

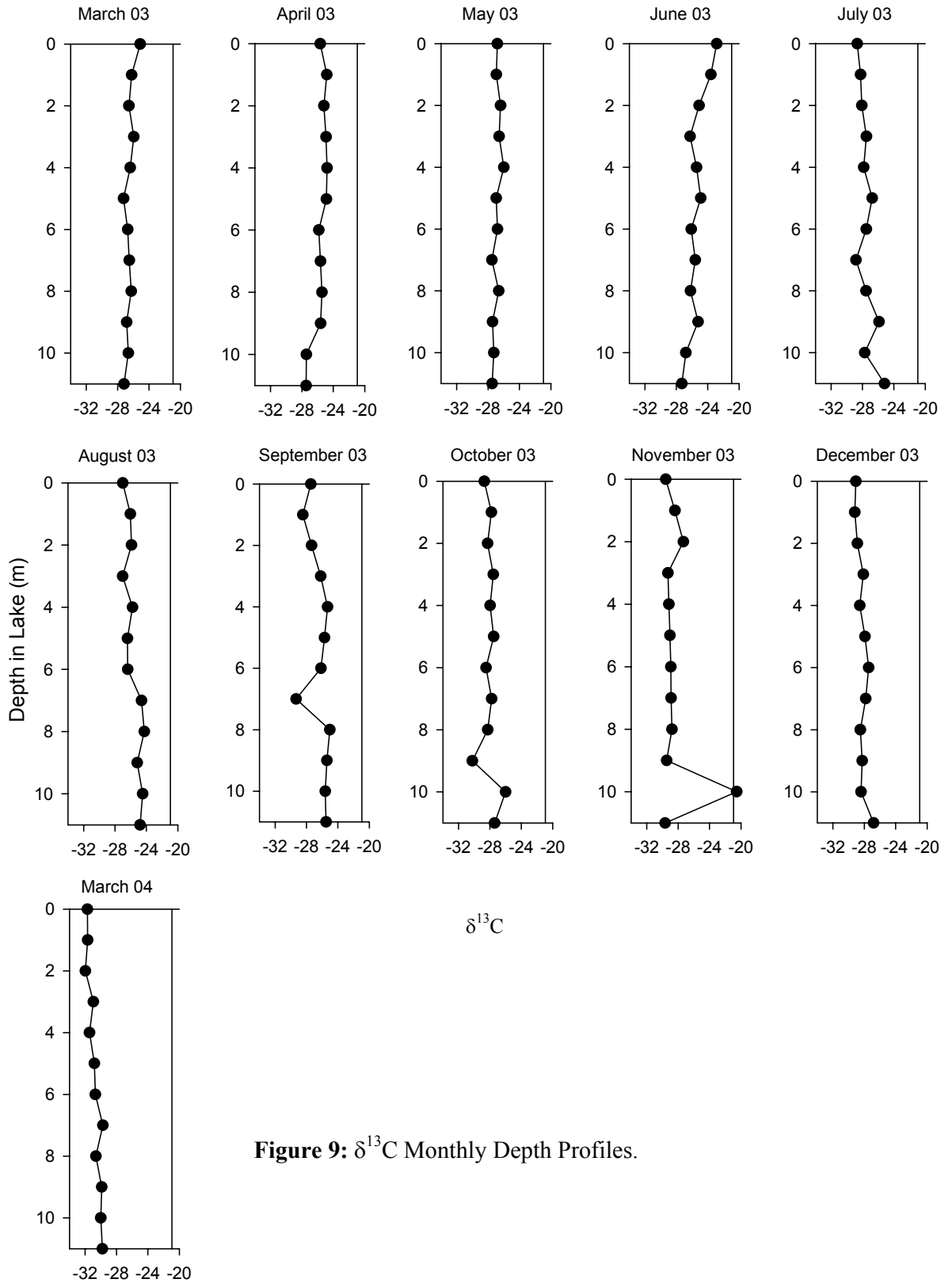


Figure 9: $\delta^{13}\text{C}$ Monthly Depth Profiles.

seasonal changes such as stratification throughout the water column, $\delta^{13}\text{C}$ showed no spatial or seasonal patterns (Figure 8). When compared to the isopleth diagrams of temperature (Figure 2), dissolved oxygen (Figure 4), or $\delta^{15}\text{N}$ (Figure 6), the spatial and seasonal $\delta^{13}\text{C}$ values of seston did not correspond to any other data collected at Cougar Lake.

CHAPTER VI

DISCUSSION

Temperature

Cougar Lake is considered monomictic (Wetzel 1983) with one period of complete mixing. In this study, the period of complete mixing in Cougar Lake begins in November and continues through March, as it did in 1999-2000 (Rieger 2003). Throughout the period of stratification the epilimnion is comprised of the top 5 meters and the water in this layer separates itself from the hypolimnion. These temperature differences at different seasons are shown in Figure 10. The vertical profiles in Figure 10 show isothermal conditions in March 2003 and the most prominent period of stratification in Cougar Lake in July 2003.

The separation of the epilimnion and the hypolimnion is observable from temperature readings at Cougar Lake as well (Appendix A). Once the surface waters are warmed at the onset of the spring season, the cool dense waters of the hypolimnion remain at the bottom of Cougar Lake until the fall turnover. As the epilimnion begins to cool and the fall turnover process begins, only then does the hypolimnion begin to mix with the epilimnion and temperatures increase. The warmest temperature at 11 meters in Cougar Lake occurred during this study at the November 2003 sampling event. The difference in water temperature between the bottom and surface waters at this time was 1.3°C, compared to a difference of 22.9°C in July 2003 during stratification. Isothermal temperatures are evidence that the fall turnover has occurred in Cougar Lake.

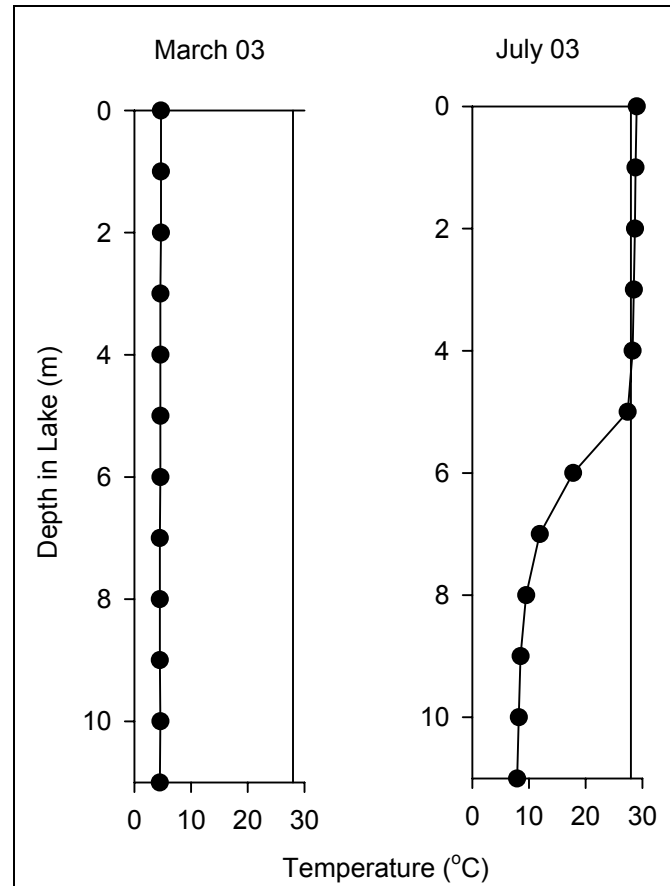


Figure 10: March and July 2003 Temperature (°C) Depth Profiles.

Dissolved Oxygen

Similar to the temperature depth profiles, dissolved oxygen readings also indicates stratified and isothermal conditions of Cougar Lake (Figure 11). Dissolved oxygen readings in the top five meters of Cougar Lake are higher than the rest of the lake throughout the stratified period between April 11, 2003 and October 17, 2003 (Appendix A) because the water in epilimnion is circulated by the wind and thus oxygen is replenished to these waters. Even though the cool dense water in the hypolimnion is capable of holding more dissolved oxygen than warm water at the surface, its oxygen supply is cut off and depleted by

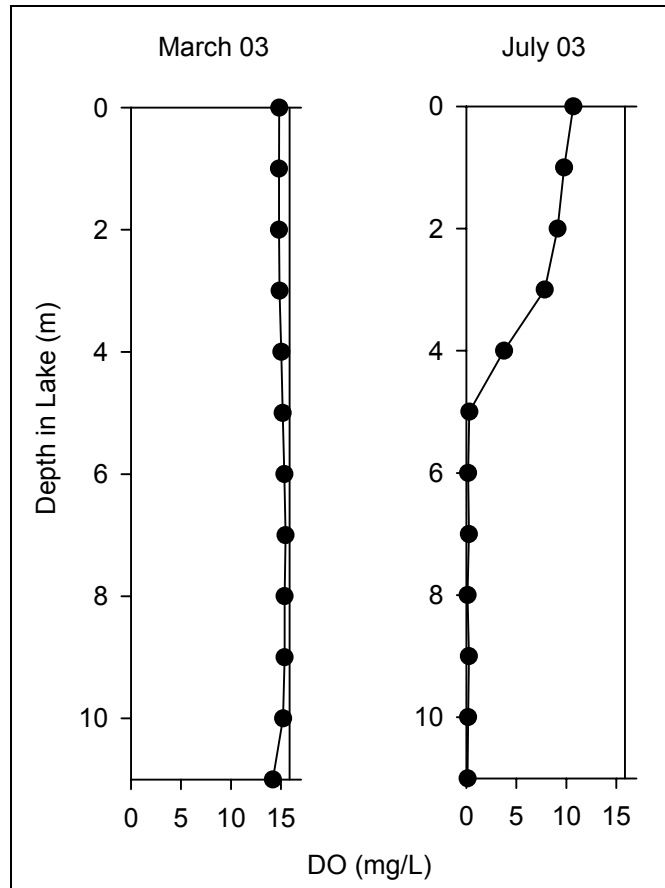


Figure 11: March and July 2003 Dissolved Oxygen (mg/L) Depth Profiles.

microbial decomposition of dead plant material. Only anaerobic bacteria are able to survive throughout the period of oxygen-deprived conditions of the hypolimnion during the summer months.

Once the temperature of the lake decreased in the fall, between the September 16, 2003 and October 17, 2003 sampling events, the water contained in the hypolimnion began to mix with the water from the epilimnion. The outcome was a gradual increase of dissolved oxygen readings in the deeper waters of Cougar Lake until December 15, 2003, when isothermal conditions once again returned.

Nitrogen Isotopes

Seston in Cougar Lake reflected seasonal changes in nitrogen isotopic composition during the study period. Relatively low $\delta^{15}\text{N}$ values corresponded with low temperature and high dissolved oxygen during isothermal conditions in April 2003. With the onset of stratification in May, $\delta^{15}\text{N}$ values in the epilimnion increased to values known to be wastewater-derived (Table 2) and corresponded with increased temperature and dissolved oxygen. This trend continued throughout the summer until the December 2003 sampling event when isothermal conditions in the water column revealed a $\delta^{15}\text{N}$ range from +4.3‰ at the surface to 6.28‰ at 11 meters, a difference of 1.98‰. This was similar to the results in March 2003 where $\delta^{15}\text{N}$ ranged from +6.21‰ at the surface to +7.02‰ at 11 meters, a difference of 0.81‰.

The increased $\delta^{15}\text{N}$ values of seston can be attributed to several factors, including assimilation of ^{15}N enriched NH_4^+ , microbial degradation, and disaggregation of sinking particulate organic mater (POM) (McCusker *et al.* 1999). These transformation processes, the $\delta^{15}\text{N}$ values of other particles contributing to the composition of seston, and the isotopic composition of the dissolved inorganic nitrogen source (McCusker *et al.* 1999) give indication of the $\delta^{15}\text{N}$ values of seston in Cougar Lake. The introduction of wastewater into Cougar Lake accounts for the high $\delta^{15}\text{N}$ values of the dissolved inorganic source (Table 2). Any transformation processes that utilize nitrogen from Cougar Lake cannot discriminate against $\delta^{15}\text{N}$ because the source of nitrogen is already depleted in ^{14}N , thus, the uptake of ^{15}N is inevitable and phytoplankton in Cougar Lake assume higher $\delta^{15}\text{N}$ values.

Compared to seston results in Grand Traverse Bay, Lake Michigan (McCusker *et al.* 1999), the highest $\delta^{15}\text{N}$ values of seston in Cougar Lake were not revealed during the onset

of stratification in the spring, but closer to the fall turnover during September 2003.

Nitrogen isotope values of seston in Grand Traverse Bay ranged from 1.7 to 11.6‰, with high $\delta^{15}\text{N}$ values recorded at 20 meters in April and May. Highest $\delta^{15}\text{N}$ readings at Cougar Lake during the entire study period were 18.48‰ and 18.7‰ at the surface and 1 meter, respectively, in September 2003. If the inorganic nitrogen source originates from the wastewater treatment facility and contributes to high $\delta^{15}\text{N}$ values, then the volume of wastewater generated throughout the year should also have a significant impact. This is especially noticeable during the September event when the fall semester begins at SIUE and the daily campus population significantly increases compared to the summer. According to the SIUE Discharge Monitoring Report submitted to the IEPA (Figure 12, Table 1), a noticeable decrease in average flow occurs between April (1,100,000 L/day) and May (720,000 L/day), and a significant increase in average flow occurs between August (over 700,000 L/day) and September 2003 (over 1.2 million L/day) when the campus population of residents and students increases dramatically.

Nitrogen input from the wastewater treatment facility is also influenced by flow, which is evident from the data provided by the Discharge Monitoring Report (Figures 12 and 13, Table 1). The first increase in average ammonia concentration during the study period coincided with the first increase in flow in April 2003. Another increase in ammonia occurred in August 2003 when the NH_3 maximum value for the month peaked at over 6 mg/L and the maximum flow increased to over 350,000 gallons/day. The first increase in April 2003 can be attributed to stratification when all incoming wastewater with high $\delta^{15}\text{N}$ values remain at or near the surface of the lake. The continuous flow of high $\delta^{15}\text{N}$ wastewater throughout the summer combined with the increase in wastewater contribution and high $\delta^{15}\text{N}$

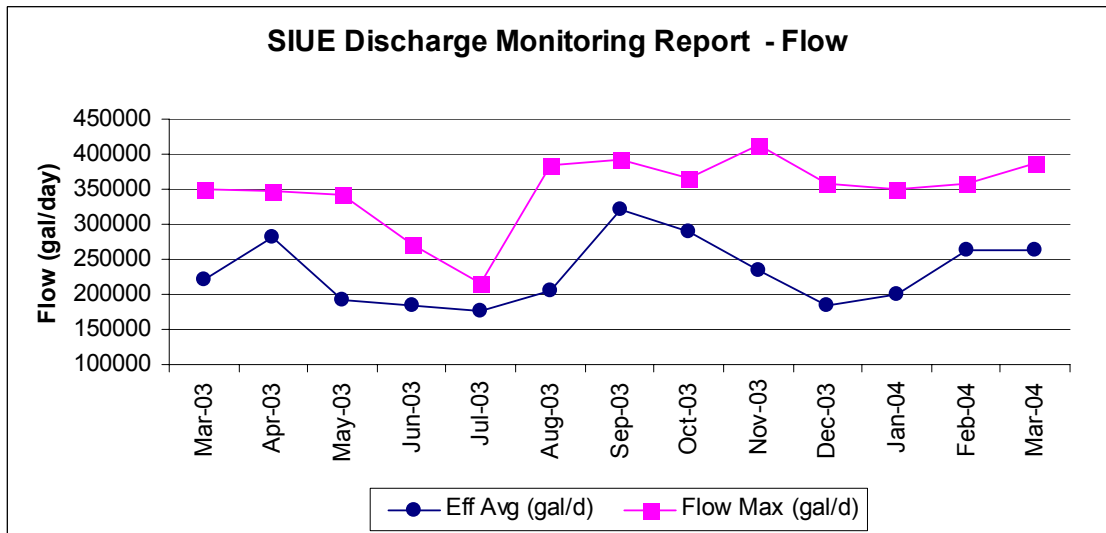


Figure 12: Monthly Flow Averages and Maximums (gallons/day) Discharged Into Cougar Lake.

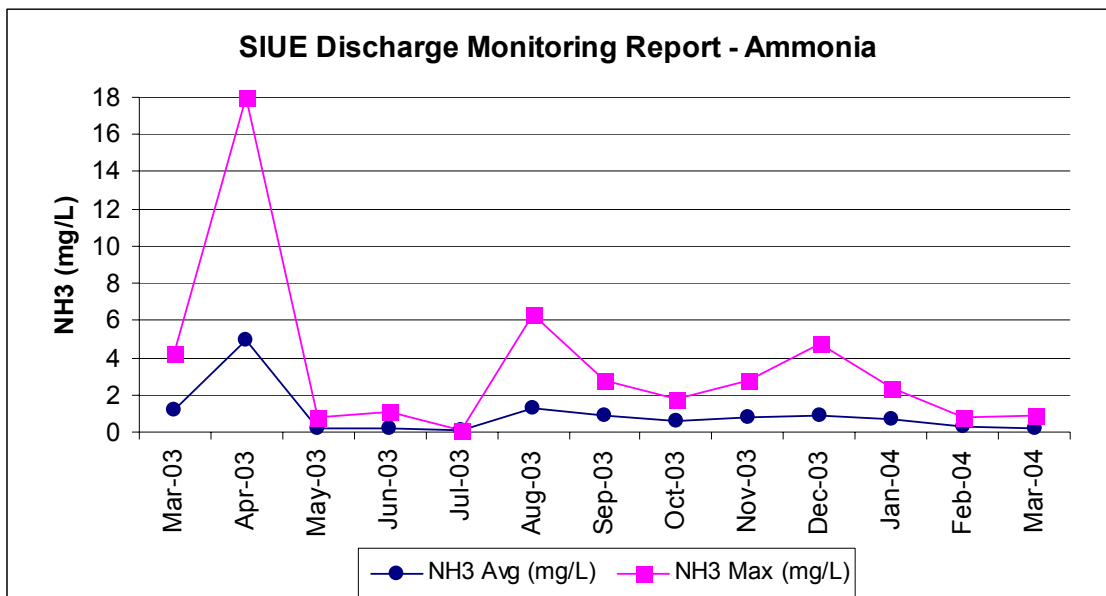


Figure 13: Monthly Ammonia Averages and Maximums (mg/L) Discharged Into Cougar Lake.

content create ideal conditions for the highest $\delta^{15}\text{N}$ values for seston in Cougar Lake during the study period.

Another source of water input in the lake is rainfall (Table 3), and a total of 130.7 centimeters of rain fell in St. Louis, MO, throughout the study period between March 2003 and March 2004 (National Weather Service 2004). Rainfall information was obtained from the National Weather Service for St. Louis, MO, because of its proximity to the campus and the information source is a major forecasting office. Over 30 centimeters of rain fell during the month of June, and only a combined 23.02 centimeters fell during the subsequent three months (July through September). The decreased rainfall combined with the decreased input from the wastewater treatment facility during July and August contributes to higher $\delta^{15}\text{N}$

Table 3: Rainfall Totals for the Study Period (National Weather Service Forecast Office, 2004)

<u>Rainfall</u>	<u>Centimeters</u>
March '03	7.00
April	10.73
May	9.93
June	30.88
July	6.28
August	6.35
September	10.38
October	7.03
November	13.35
December	5.85
January '04	9.93
February	2.13
March	10.90

signatures of seston during those months. As nitrate becomes limiting, the preferential removal of ^{14}N no longer occurs and phytoplankton growing in this area no longer discriminate against ^{15}N , eventually leading to a $\delta^{15}\text{N}$ similar to nitrate (Ostrom *et al.* 1997). Since the major source of water and nitrogen input to Cougar Lake during the summer originates from the wastewater treatment facility and the inorganic nitrogen coming from this effluent contains high $\delta^{15}\text{N}$ values, the $\delta^{15}\text{N}$ values of seston are much higher in the late summer than the rest of the year and resemble $\delta^{15}\text{N}$ values of the major nitrogen source.

The extended period of stratification through the fall until November when stratification finally breaks also contributes to the high $\delta^{15}\text{N}$ values. The temperature in the top 4 meters remained above 18°C until October when thermal stratification began to break down. During this time algae and phytoplankton have optimal conditions to continue utilizing the inorganic nitrogen source available from the wastewater treatment facility. In a previous study (Rosen 1978), thermal stratification began to break down as early as September, although temperatures in the top 5 meters exceeded 18°C until the end of October and complete mixing with the bottom waters could occur at this time. The delayed fall turnover and subsequent mixing of the lake water provides optimal conditions for nitrogen enrichment processes such as assimilation of ^{15}N -enriched NH_4^+ and microbial degradation (McCusker *et al.* 1999).

Seasonal changes in $\delta^{15}\text{N}$ values for seston in Cougar Lake are similar to those in Lake Michigan (McCusker *et al.* 1999). McCusker *et al.* (1999) found that the uptake of ^{15}N -enriched NH_4^+ influences high $\delta^{15}\text{N}$ values for seston. The combination of high $\delta^{15}\text{N}$ in the surface 20 meters and a decrease in NH_4^+ concentrations from April to May are consistent with uptake of NH_4^+ by phytoplankton in Lake Michigan. Similar conditions existed in Cougar Lake between April and May 2003. Increased $\delta^{15}\text{N}$ values of seston

(Appendix A) were recorded (10.56‰ to 13.02‰ at 0 meters) from April 11, 2003 to May 14, 2003 and a significant decrease in average ammonia (Table 1) was discharged from the wastewater treatment facility into Cougar Lake (from 4.99 mg/L in April to 0.16 mg/L in May). Therefore, the uptake of NH_4^+ by phytoplankton in Cougar Lake is also evident between the months of April and May and impacts $\delta^{15}\text{N}$ of seston.

Carbon Isotopes

Based on the analysis of carbon isotopes of seston in Cougar Lake, no patterns of productivity changes can be seen in Cougar Lake (Figure 8). Based on Figure 8, there are no distinct spatial or temporal distributions of $\delta^{13}\text{C}$. Phytoplankton in Cougar Lake appear to discriminate normally against ^{13}C and photosynthesis is not carbon-limited throughout the study period. If carbon-limited photosynthesis occurs, the $\delta^{13}\text{C}$ values of seston would reflect higher (less negative) values than the range of -20.55‰ to -31.97‰ in this study.

The depth profiles of $\delta^{13}\text{C}$ from each sampling event (Figure 9) indicate slight changes with depth. From the March 2003 sampling event to May 2003, $\delta^{13}\text{C}$ values appear to be similar throughout the entire water column. Starting in the summer months of July and August and into the September sampling event, $\delta^{13}\text{C}$ values are slightly elevated near the bottom of Cougar Lake. High $\delta^{13}\text{C}$ values for seston (ranging from -23.9‰ to -26.0‰) in Grand Traverse Bay, Michigan, at depths of 60 meters and below were related to $\delta^{13}\text{C}$ of sediment, and resuspension of sedimentary material in late summer contributed to those high values (McCusker *et al.*, 1999). This can also explain the large peak in $\delta^{13}\text{C}$ at 10 meters in November 2003, where $\delta^{13}\text{C}$ was -20.55‰.

The $\delta^{13}\text{C}$ values of seston in Cougar Lake also indicate that photosynthesis in the water column is not carbon limited. Terrestrial plants and phytoplankton in and around

Cougar Lake undergo C_3 photosynthesis, which has a $\delta^{13}C$ range of -20‰ to -35‰ (Wang *et al.* 1998). The wide range is attributed to the C isotopic composition of the atmosphere, which is currently -8‰, and the tendency for C_3 plants to have ^{13}C -enriched values in water-stressed conditions (Wang *et al.* 1998). Plants uptake CO_2 from the atmosphere and phytoplankton uptake CO_2 directly from the water, thus the $\delta^{13}C$ of seston is enriched (less negative) when CO_2 is limited in the water column. Any dead or decaying plant material included and analyzed as seston in the water column portrays $\delta^{13}C$ values based on the type of photosynthesis being utilized, which is C_3 photosynthesis in Cougar Lake (Table 2).

CHAPTER VII

CONCLUSIONS

Stable isotope analysis of seston in Cougar Lake on the campus of Southern Illinois University Edwardsville is a significant indicator of both physical limnology and biological processes throughout the water column. The stable nitrogen isotopes varied the most throughout the study period and depth profiles compared to stable carbon isotopes. When compared to the isopleth diagrams of temperature and dissolved oxygen, the $\delta^{15}\text{N}$ values of seston appeared to have similar seasonal patterns throughout the study period, where high values were contained in the hypolimnion during stratification and relatively lower $\delta^{15}\text{N}$ values were prevalent during times of complete mixing in the water column. Conversely, stable carbon isotopic analysis of seston revealed only the primary method of photosynthesis, which is evidence of C_3 photosynthesis in Cougar Lake.

Important seasonal influences on the stable isotope geochemistry in Cougar Lake, such as temperature, dissolved oxygen, rainfall, and other inputs in the lake were discovered during the study period. Although rainfall $\delta^{15}\text{N}$ values could not be obtained, runoff from rainfall events can contribute to $\delta^{15}\text{N}$ values in seston based on rainfall amounts and types of nitrogen contributing to the runoff, such as guano left along the shores from the population of geese on campus throughout the year. It was determined that the wastewater treatment facility serving the campus was the main contributor to the increased $\delta^{15}\text{N}$ values of seston because the highest values occurred at the same time as a peak in average flow per day. Wastewater discharged into the lake already has a high $\delta^{15}\text{N}$ signature, and the increased volume discharged corresponds to the peaks of high $\delta^{15}\text{N}$ of seston in the lake during the study period.

Contrary to what was hypothesized, the highest $\delta^{15}\text{N}$ values were revealed later in the stratification period, rather than at its onset. Also, carbon isotopes did not change throughout the study period with changing productivity levels as was first expected. Direct impact of the use of lake water for the heating/refrigeration plant could not be related to changing isotopic composition or delayed stratification, which indicates no significant problems with the plant's use of lake water.

REFERENCES

- Bernasconi, Stefano M., Alberto Barbieri, Marco Simona, 1997. Carbon and nitrogen isotope variations in sedimenting organic matter in Lake Lugano. *Limnol. Oceanogr.* 42 (8), 1755-1765.
- Brady, Katherine. 1999. Phosphorus and silica cycling in a Midwestern hypereutrophic reservoir. Thesis. Graduate School of Southern Illinois University Edwardsville.
- Cole, Marci L., Ivan Valiela, Kevin D. Kroeger, Gabrielle L. Tomaskyh, Just Cebrian, Cathleen Wigand, Richard A. McKinney, Sara P. Grady, Maria Helena Carvalho da Silva, 2004. Assessment of a $\delta^{15}\text{N}$ isotopic method to indicate anthropogenic eutrophication in aquatic ecosystems. *J. Environ. Qual.* 33 (1), 124-132.
- Deming, Tricia. 2000. The cycling of iron, manganese, and sulfide in Cougar (Tower) Lake, Madison County, Illinois. Thesis. Graduate School of Southern Illinois University Edwardsville.
- Goericke, R., J. P. Montoya, B. Fry. 1994. Physiology of isotopic fractionation in algae and cyanobacteria. *Stable Isotopes in Ecology and Environmental Science*. Blackwell, Boston. 187-221.
- Graham, M.C., M.A. Eaves, J.G. Farmer, J. Dobson, A.E. Fallick, 2001. A study of carbon and nitrogen stable isotope and elemental ratios as potential indicators of source and fate of organic matter in sediments of the Forth Estuary, Scotland. *Estuarine, Coastal, and Shelf Science.* 52, 375-380.
- Griggs, Erin M., Lee R. Kump, J.K. Bohlke. 2003. The fate of wastewater-derived nitrate in the subsurface of the Florida Keys: Key Colony Beach, Florida. *Estuarine, Coastal, and Shelf Science.* 58, 517-539.
- Guo, Chu. 2002. Accumulation of copper algacide in the sediment of Cougar Lake, a small Illinois Reservoir. Thesis. Graduate School of Southern Illinois University Edwardsville.
- Hodell, David A., Claire L. Schelske, 1998. Production, sedimentation, and isotopic composition of organic matter in Lake Ontario. *Limnol. Oceanogr.* 43(2), 200-214.
- Huang, S.-C., D.A. Kreeger, R.I.E. Newell. 2003. Seston available as a food source for the ribbed mussel (*Geukensia demissa*) in a North American, mid-Atlantic saltmarsh. *Estuarine, Coastal, and Shelf Science.* 56, 561-571.
- Illinois Environmental Protection Agency. 2004. Discharge Monitoring Report for Southern Illinois University Edwardsville Outfall 0010.

- Kendall, Carol, 1998. Tracing Nitrogen Sources and Cycling in Catchments. Isotope Tracers in Catchment Hydrology. Elsevier, Amsterdam. 519-576.
- Kendall, Carol, Eric A. Caldwell, 1998. Fundamentals of Isotope Geochemistry. Isotope Tracers in Catchment Hydrology. Elsevier, Amsterdam. 51-86.
- Lajeone, Larry J. 1972. The physical limnology of Tower Lake, Madison County, Illinois. Thesis. Graduate School of Southern Illinois University, Edwardsville.
- Lehmann, Moritz F., Peter Reichert, Stefano M. Bernasconi, Alberto Barbieri, Judith A. McKenzie, 2003. Modelling nitrogen and oxygen isotope fractionation during denitrification in a lacustrine redox-transition zone. *Geochimica et Cosmochimica Acta* 67 (14), 2529-2542.
- Macko, S. A., N.E. Ostrom, 1994. Pollution studies using stable isotopes. Stable Isotopes in Ecology and Environmental Science. Blackwell, Boston. 45-62.
- McClelland, James W., Ivan Valiela, 1998. Linking nitrogen in estuarine producers to land-derived sources. *Limnol. Oceanogr.* 43 (4), 577-585.
- McCusker, Eileen M., Peggy H. Ostrom, Nathaniel E. Ostrom, Jeffery D. Jeremiason, Joel E. Baker, 1999. Seasonal variation in biogeochemical cycling of seston in Grand Traverse Bay, Lake Michigan. *Organic Geochemistry* 30, 1543-1557.
- National Weather Service Forecast Office, St. Louis, MO. Climatology and Weather Records. [Online] Available <http://www.crh.noaa.gov/lx/climate.php>, November 15, 2004.
- Ostrom, Nathaniel E., Stephen A. Macko, Don Deibel, Raymond J. Thompson, 1997. Seasonal variation in the stable carbon and nitrogen isotope biogeochemistry of a coastal cold ocean environment. *Geochimica et Cosmochimica Acta* 61 (14), 2929-2942.
- Ostrom, Nathaniel E., David T. Long, Emily M. Bell, Tina Beals, 1998. The origin and cycling of particulate sedimentary organic matter and nitrate in Lake Superior. *Chemical Geology* 152, 13-28.
- Rieger, Chad. 2003. A Phosphorus Budget for Cougar Lake in Madison County, Illinois. Thesis. Southern Illinois University Edwardsville.
- Rosen, Martin G. 1978. A limnological survey of Tower Lake, Madison County, Illinois, with special reference to the presence of artificial aeration. Thesis. Southern Illinois University Edwardsville.
- Thornton, S. F., J. McManus, 1994. Application of organic carbon and nitrogen stable isotope and C/N ratios as source indicators of organic matter provenance in estuarine

systems: Evidence from the Tay Estuary, Scotland. *Estuarine, Coastal and Shelf Science* 38, 219-233.

Vuorio, K., A.M. Ventela, J. Sipura, M. Tarvainen, M. Meili, J. Sarvala, J. 2002. Stable carbon and nitrogen isotopes in lake plankton and seston: variability among 10 fractions, two seasons, and two lakes. *Verh. Internat. Verein. Limnol.* 28, 1396-1399.

Wang, Yang, Thomas G. Huntington, Laurie J. Osher, Leonard I. Wassenaar, Susan E. Trumbore, Ronald G. Amundson, Jennifer W. Harden, Diane M. McKnight, Sherry L. Schiff, George R. Aiken, W. Berry Lyons, Ramon O. Aravena, Jill S. Baron. 1998. Carbon Cycling in Terrestrial Environments. Isotope Tracers in Catchment Hydrology. Elsevier, Amsterdam, 577-610.

Wetzel, Robert G. 1983. Limnology. Philadelphia, Saunders.

APPENDIX A

Raw Data Collected From Cougar Lake

Table A-1: Seston Data, Dissolved Oxygen, and Temperature Readings Collected at Cougar Lake (J Date = Julian Date; Depth = meters; DO = dissolved oxygen (mg/L); Temp. = Temperature in °C; C:N = % Carbon to % Nitrogen ratio; ND=not detected)

<u>J Date</u>	<u>Date</u>	<u>Depth</u>	<u>$\delta^{15}\text{N}$</u>	<u>$\delta^{13}\text{C}$</u>	<u>DO</u>	<u>Temp.</u>	<u>%C</u>	<u>%N</u>	<u>C:N</u>
70	3/11/2003	0	6.21	-25.13	14.82	4.7	2.05	0.28	7.32
70	3/11/2003	1	6.22	-26.22	14.81	4.7	2.2	0.3	7.33
70	3/11/2003	2	7.32	-26.56	14.8	4.7	2.73	0.32	8.53
70	3/11/2003	3	6.94	-25.95	14.87	4.6	2.13	0.3	7.10
70	3/11/2003	4	6.61	-26.39	15.04	4.6	2.09	0.32	6.53
70	3/11/2003	5	6.46	-27.24	15.17	4.6	1.93	0.29	6.66
70	3/11/2003	6	1.87	-26.72	15.34	4.6	2.01	0.22	9.14
70	3/11/2003	7	8.68	-26.52	15.45	4.5	2.24	0.31	7.23
70	3/11/2003	8	7.16	-26.27	15.35	4.5	1.95	0.3	6.50
70	3/11/2003	9	6.9	-26.85	15.36	4.5	2.34	0.28	8.36
70	3/11/2003	10	6.59	-26.65	15.2	4.6	1.62	0.24	6.75
70	3/11/2003	11	7.02	-27.18	14.18	4.5	1.97	0.27	7.30
70	3/11/2003	Blank	ND	-27.72			1.47	ND	
101	4/11/2003	0	10.56	-25.66	8.97	18	1.39	0.14	9.93
101	4/11/2003	1	10.77	-24.82	9.2	17.1	0.79	0.09	8.78
101	4/11/2003	2	11.43	-25.21	9.5	16	0.98	0.1	9.80
101	4/11/2003	3	11.3	-24.92	9.67	15.3	0.94	0.13	7.23
101	4/11/2003	4	11.83	-24.8	9.87	14.7	0.97	0.13	7.46
101	4/11/2003	5	11.43	-24.88	10.07	14	1.04	0.1	10.40
101	4/11/2003	6	12.12	-25.86	10.28	13.3	1.06	0.11	9.64
101	4/11/2003	7	11.81	-25.64	10.55	12.4	1.1	0.09	12.22
101	4/11/2003	8	11.09	-25.45	10.75	11.8	0.96	0.08	12.00
101	4/11/2003	9	10.91	-25.62	12.51	10.8	0.92	0.08	11.50
101	4/11/2003	10	9.85	-27.43	13.02	9.6	1.28	0.12	10.67
101	4/11/2003	11	7.44	-27.47	13.6	8.9	0.94	0.11	8.55
101	4/11/2003	Blank	ND	-24.36			0.53	ND	
134	5/14/2003	0	13.02	-26.82	11.06	19.8	1.47	0.17	8.65
134	5/14/2003	1	13.25	-26.94	11.04	20	1.48	0.17	8.71
134	5/14/2003	2	10.93	-26.41	10.4	19.9	1.53	0.13	11.77
134	5/14/2003	3	12.04	-26.59	7.94	19.1	1.56	0.17	9.18
134	5/14/2003	4	10.32	-26	6.15	18.2	1.07	0.1	10.70
134	5/14/2003	5	10.44	-26.96	1.13	15.1	1.45	0.12	12.08
134	5/14/2003	6	6.87	-26.81	0.12	9.8	1.05	0.07	15.00
134	5/14/2003	7	6.23	-27.52	0.05	8.1	1.06	0.08	13.25
134	5/14/2003	8	7.25	-26.64	0.15	7.3	0.99	0.09	11.00
134	5/14/2003	9	5.57	-27.43	0.04	6.9	0.99	0.07	14.14
134	5/14/2003	10	6.48	-27.28	0.03	6.7	0.91	0.08	11.38

<u>J Date</u>	<u>Date</u>	<u>Depth</u>	<u>$\delta^{15}\text{N}$</u>	<u>$\delta^{13}\text{C}$</u>	<u>DO</u>	<u>Temp.</u>	<u>%C</u>	<u>%N</u>	<u>C:N</u>
134	5/14/2003	11	6.1	-27.48	0.03	6.6	1.03	0.07	14.71
134	5/14/2003	Blank	ND	-27.09			0.84	ND	
169	6/18/2003	0	9.73	-22.86	9.6	28.3	1	0.07	14.29
169	6/18/2003	1	11.17	-23.6	9.46	27.6	3.31	0.2	16.55
169	6/18/2003	2	11.87	-25.09	8.12	24.9	1.99	0.15	13.27
169	6/18/2003	3	11.12	-26.25	2.64	22.7	2.02	0.19	10.63
169	6/18/2003	4	14.63	-25.43	0.29	20.9	1.31	0.12	10.92
169	6/18/2003	5	9.16	-24.89	0.1	18.6	1.1	0.07	15.71
169	6/18/2003	6	2.15	-26.11	0.07	13.7	1.43	0.08	17.88
169	6/18/2003	7	8.1	-25.58	0.06	9.5	0.97	0.07	13.86
169	6/18/2003	8	7.84	-26.2	0.03	6.4	1.07	0.07	15.29
169	6/18/2003	9	7.4	-25.23	0.07	7.6	0.78	0.07	11.14
169	6/18/2003	10	7.37	-26.78	0.05	7.3	1.48	0.07	21.14
169	6/18/2003	11	7.99	-27.3	0.05	7.2	1.42	0.08	17.75
169	6/18/2003	Blank	ND	-28.00			1.78	ND	
203	7/22/2003	0	10.03	-28.67	10.7	29	1.7	0.16	10.63
203	7/22/2003	1	9.8	-28.24	9.77	28.8	1.35	0.15	9.00
203	7/22/2003	2	9.54	-28.08	9.13	28.7	1.9	0.15	12.67
203	7/22/2003	3	9.3	-27.49	7.84	28.5	1.08	0.13	8.31
203	7/22/2003	4	10.25	-27.84	3.77	28.3	1.38	0.09	15.33
203	7/22/2003	5	8.54	-26.75	0.29	27.4	0.89	0.07	12.71
203	7/22/2003	6	6.54	-27.52	0.18	17.8	1.65	0.08	20.63
203	7/22/2003	7	5.88	-28.83	0.25	11.9	2.37	0.08	29.63
203	7/22/2003	8	5.9	-27.55	0.12	9.5	1.55	0.08	19.38
203	7/22/2003	9	6.54	-25.88	0.25	8.5	0.97	0.08	12.13
203	7/22/2003	10	5.54	-27.72	0.19	8.2	1.84	0.08	23.00
203	7/22/2003	11	5.26	-25.19	0.12	7.9	0.93	0.1	9.30
203	7/22/2003	Blank	ND	-26.71			0.89	ND	
224	8/12/2003	0	9.22	-27.02	10.01	28.3	0.6	0.08	7.50
224	8/12/2003	1	9.08	-26.04	10.08	28.3	0.65	0.09	7.22
224	8/12/2003	2	9.06	-25.9	9.83	28.3	0.64	0.09	7.11
224	8/12/2003	3	8.78	-27.04	9.38	28.2	0.61	0.08	7.63
224	8/12/2003	4	8.5	-25.78	8.1	28.1	0.59	0.07	8.43
224	8/12/2003	5	7.75	-26.43	2.8	27.6	0.49	0.05	9.80
224	8/12/2003	6	6.3	-26.39	0.52	20.3	0.63	0.07	9.00
224	8/12/2003	7	5.23	-24.62	0.64	11.9	0.67	0.08	8.38
224	8/12/2003	8	5.51	-24.28	0.4	9.8	0.65	0.08	8.13
224	8/12/2003	9	4.35	-25.18	0.34	8.6	0.75	0.09	8.33
224	8/12/2003	10	4.38	-24.48	0.14	8	0.78	0.1	7.80
224	8/12/2003	11	4.55	-24.81	0.05	7.9	0.91	0.13	7.00
224	8/12/2003	Blank	0.19	-25.44			0.27	0.82	

<u>J Date</u>	<u>Date</u>	<u>Depth</u>	<u>$\delta^{15}\text{N}$</u>	<u>$\delta^{13}\text{C}$</u>	<u>DO</u>	<u>Temp.</u>	<u>%C</u>	<u>%N</u>	<u>C:N</u>
259	9/16/2003	0	18.48	-27.45	15.67	26.5	1.28	0.15	8.53
259	9/16/2003	1	18.7	-28.48	13.88	26	0.98	0.16	6.13
259	9/16/2003	2	15.24	-27.36	13.66	25.7	1.36	0.09	15.11
259	9/16/2003	3	12.56	-26.18	9.95	24.6	1.17	0.07	16.71
259	9/16/2003	4	9.1	-25.32	6.85	24.4	1.09	0.04	27.25
259	9/16/2003	5	8.39	-25.71	6.07	24.3	1.12	0.04	28.00
259	9/16/2003	6	7.15	-26.15	1.51	23.7	1.14	0.05	22.80
259	9/16/2003	7	4.27	-29.33	0.15	18.8	1.21	0.07	17.29
259	9/16/2003	8	5.18	-25.03	0.11	11.9	1.08	0.07	15.43
259	9/16/2003	9	5.29	-25.39	0.09	10.3	1.02	0.08	12.75
259	9/16/2003	10	5.07	-25.61	0.07	8.8	0.99	0.08	12.38
259	9/16/2003	11	3.75	-25.5	0.08	8.4	1.1	0.12	9.17
259	9/16/2003	Blank	ND	ND			ND	ND	
290	10/17/2003	0	9.46	-28.71	7.11	18.1	0.98	0.08	12.25
290	10/17/2003	1	8.44	-27.81	6.56	18.1	0.97	0.07	13.86
290	10/17/2003	2	8.72	-28.3	6.3	17.9	0.94	0.06	15.67
290	10/17/2003	3	7.37	-27.58	6.09	17.8	0.94	0.06	15.67
290	10/17/2003	4	11.32	-27.97	5.96	17.8	0.92	0.04	23.00
290	10/17/2003	5	12	-27.53	5.92	17.8	1.01	0.06	16.83
290	10/17/2003	6	11.03	-28.49	5.87	17.8	0.95	0.04	23.75
290	10/17/2003	7	12.46	-27.79	5.38	17.7	0.94	0.04	23.50
290	10/17/2003	8	9.99	-28.28	3.6	17	0.94	0.05	18.80
290	10/17/2003	9	5.71	-30.25	1.07	13	0.94	0.07	13.43
290	10/17/2003	10	7.25	-26.02	0.53	10	0.94	0.07	13.43
290	10/17/2003	11	6.11	-27.41	0.35	9.6	0.99	0.09	11.00
290	10/17/2003	Blank	ND	ND			ND	ND	
316	11/12/2003	0	8.11	-29.59	5.84	13.6	1.04	0.04	26.00
316	11/12/2003	1	9.94	-28.42	5.24	13.5	1.01	0.04	25.25
316	11/12/2003	2	9.02	-27.34	5.11	13.5	1.17	0.04	29.25
316	11/12/2003	3	5.47	-29.32	4.94	13.2	1.11	0.05	22.20
316	11/12/2003	4	4.86	-29.19	4.3	12.8	1.08	0.05	21.60
316	11/12/2003	5	5.4	-29.04	3.91	12.7	1.01	0.05	20.20
316	11/12/2003	6	5.28	-28.93	3.74	12.5	1.12	0.06	18.67
316	11/12/2003	7	5.61	-28.9	3.46	12.4	1.24	0.06	20.67
316	11/12/2003	8	5.64	-28.8	3.21	12.4	1.07	0.06	17.83
316	11/12/2003	9	5.7	-29.48	3.71	12.4	1.1	0.07	15.71
316	11/12/2003	10	5.17	-20.55	3.36	12.3	0.06	0.06	1.00
316	11/12/2003	11	4.17	-29.66	3.14	12.3	1.19	0.06	19.83
316	11/12/2003	Blank	ND	ND			ND	ND	
349	12/15/2003	0	4.3	-29.08	9.01	5.2	0.46	0.05	9.20
349	12/15/2003	1	4.63	-29.21	8.94	5.2	0.54	0.06	9.00
349	12/15/2003	2	4.37	-28.88	8.94	5.2	0.9	0.1	9.00

<u>J Date</u>	<u>Date</u>	<u>Depth</u>	<u>$\delta^{15}\text{N}$</u>	<u>$\delta^{13}\text{C}$</u>	<u>DO</u>	<u>Temp.</u>	<u>%C</u>	<u>%N</u>	<u>C:N</u>
349	12/15/2003	3	4.83	-28.12	8.94	5.2	0.62	0.08	7.75
349	12/15/2003	4	5.26	-28.58	8.94	5.2	0.54	0.06	9.00
349	12/15/2003	5	5.47	-27.93	8.97	5.3	0.49	0.06	8.17
349	12/15/2003	6	4.90	-27.46	8.97	5.3	0.5	0.06	8.33
349	12/15/2003	7	4.67	-27.82	8.98	5.3	0.54	0.07	7.71
349	12/15/2003	8	5.04	-28.52	9.01	5.3	0.52	0.07	7.43
349	12/15/2003	9	5.04	-28.27	9.01	5.3	0.58	0.06	9.67
349	12/15/2003	10	4.94	-28.42	9.01	5.3	0.52	0.06	8.67
349	12/15/2003	11	6.28	-26.82	9	5.2	0.55	0.07	7.86
349	12/15/2003	Blank	ND	-24.74			0.17	ND	
427	3/2/2004	0	12.93	-31.72	12.6	7.7	1.19	0.18	6.61
427	3/2/2004	1	13.36	-31.69	13.3	7.5	1.39	0.23	6.04
427	3/2/2004	2	13.34	-31.97	13.59	7.3	1.39	0.22	6.32
427	3/2/2004	3	13.49	-30.97	13.55	6.9	1.14	0.19	6.00
427	3/2/2004	4	13.80	-31.45	13.24	6.7	1.13	0.19	5.95
427	3/2/2004	5	13.44	-30.83	13.35	6.6	1.01	0.16	6.31
427	3/2/2004	6	12.98	-30.71	13.69	6.5	0.99	0.14	7.07
427	3/2/2004	7	12.59	-29.74	13.8	6.5	0.91	0.14	6.50
427	3/2/2004	8	13.80	-30.62	14.06	6.2	0.84	0.13	6.46
427	3/2/2004	9	12.22	-29.89	14.35	6	0.78	0.12	6.50
427	3/2/2004	10	13.48	-30.02	14.42	5.9	0.78	0.12	6.50
427	3/2/2004	11	13.29	-29.81	13.8	5.7	0.75	0.11	6.82
427	3/2/2004	Blank	ND	-28.01			0.32	ND	

Table A-2: Seston Data, Dissolved Oxygen, and Temperature Readings Collected at Cougar Lake by Variable

Temperature (°C)											
	<u>3/03</u>	<u>4/03</u>	<u>5/03</u>	<u>6/03</u>	<u>7/03</u>	<u>8/03</u>	<u>9/03</u>	<u>10/03</u>	<u>11/03</u>	<u>12/03</u>	<u>3/04</u>
0	4.7	18	19.8	28.3	29	28.3	26.5	18.1	13.6	5.2	7.7
1	4.7	17.1	20	27.6	28.8	28.3	26	18.1	13.5	5.2	7.5
2	4.7	16	19.9	24.9	28.7	28.3	25.7	17.9	13.5	5.2	7.3
3	4.6	15.3	19.1	22.7	28.5	28.2	24.6	17.8	13.2	5.2	6.9
4	4.6	14.7	18.2	20.9	28.3	28.1	24.4	17.8	12.8	5.2	6.7
5	4.6	14	15.1	18.6	27.4	27.6	24.3	17.8	12.7	5.3	6.6
6	4.6	13.3	9.8	13.7	17.8	20.3	23.7	17.8	12.5	5.3	6.5
7	4.5	12.4	8.1	9.5	11.9	11.9	18.8	17.7	12.4	5.3	6.5
8	4.5	11.8	7.3	6.4	9.5	9.8	11.9	17	12.4	5.3	6.2
9	4.5	10.8	6.9	7.6	8.5	8.6	10.3	13	12.4	5.3	6
10	4.6	9.6	6.7	7.3	8.2	8	8.8	10	12.3	5.3	5.9
11	4.5	8.9	6.6	7.2	7.9	7.9	8.4	9.6	12.3	5.2	5.7

Dissolved O₂ (mg/L)											
	<u>3/03</u>	<u>4/03</u>	<u>5/03</u>	<u>6/03</u>	<u>7/03</u>	<u>8/03</u>	<u>9/03</u>	<u>10/03</u>	<u>11/03</u>	<u>12/03</u>	<u>3/04</u>
0	14.82	8.97	11.06	9.6	10.7	10.01	15.67	7.11	5.84	9.01	12.6
1	14.81	9.2	11.04	9.46	9.77	10.08	13.88	6.56	5.24	8.94	13.3
2	14.8	9.5	10.4	8.12	9.13	9.83	13.66	6.3	5.11	8.94	13.59
3	14.87	9.67	7.94	2.64	7.84	9.38	9.95	6.09	4.94	8.94	13.55
4	15.04	9.87	6.15	0.29	3.77	8.1	6.85	5.96	4.3	8.94	13.24
5	15.17	10.07	1.13	0.1	0.29	2.8	6.07	5.92	3.91	8.97	13.35
6	15.34	10.28	0.12	0.07	0.18	0.52	1.51	5.87	3.74	8.97	13.69
7	15.45	10.55	0.05	0.06	0.25	0.64	0.15	5.38	3.46	8.98	13.8
8	15.35	10.75	0.15	0.03	0.12	0.4	0.11	3.6	3.21	9.01	14.06
9	15.36	12.51	0.04	0.07	0.25	0.34	0.09	1.07	3.71	9.01	14.35
10	15.2	13.02	0.03	0.05	0.19	0.14	0.07	0.53	3.36	9.01	14.42
11	14.18	13.6	0.03	0.05	0.12	0.05	0.08	0.35	3.14	9	13.8

$\delta^{15}\text{N}$

	<u>3/03</u>	<u>4/03</u>	<u>5/03</u>	<u>6/03</u>	<u>7/03</u>	<u>8/03</u>	<u>9/03</u>	<u>10/03</u>	<u>11/03</u>	<u>12/03</u>	<u>3/04</u>
0	6.21	10.56	13.02	9.73	10.03	9.22	18.48	9.46	8.11	4.3	12.93
1	6.22	10.77	13.25	11.17	9.8	9.08	18.7	8.44	9.94	4.63	13.36
2	7.32	11.43	10.93	11.87	9.54	9.06	15.24	8.72	9.02	4.37	13.34
3	6.94	11.3	12.04	11.12	9.3	8.78	12.56	7.37	5.47	4.83	13.49
4	6.61	11.83	10.32	14.63	10.25	8.5	9.1	11.32	4.86	5.26	13.8
5	6.46	11.43	10.44	9.16	8.54	7.75	8.39	12	5.4	5.47	13.44
6	*7.57	12.12	6.87	*8.63	6.54	6.3	7.15	11.03	5.28	4.9	12.98
7	8.68	11.81	6.23	8.1	5.88	5.23	4.27	12.46	5.61	4.67	12.59
8	7.16	11.09	7.25	7.84	5.9	5.51	5.18	9.99	5.64	5.04	13.8
9	6.9	10.91	5.57	7.4	6.54	4.35	5.29	5.71	5.7	5.04	12.22
10	6.59	9.85	6.48	7.37	5.54	4.38	5.07	7.25	5.17	4.94	13.48
11	7.02	7.44	6.1	7.99	5.26	4.55	3.75	6.11	4.17	6.28	13.29

 $\delta^{13}\text{C}$

	<u>3/03</u>	<u>4/03</u>	<u>5/03</u>	<u>6/03</u>	<u>7/03</u>	<u>8/03</u>	<u>9/03</u>	<u>10/03</u>	<u>11/03</u>	<u>12/03</u>	<u>3/04</u>
0	-25.13	-25.66	-26.82	-22.86	-28.67	-27.02	-27.45	-28.71	-29.59	-29.08	-31.72
1	-26.22	-24.82	-26.94	-23.6	-28.24	-26.04	-28.48	-27.81	-28.42	-29.21	-31.69
2	-26.56	-25.21	-26.41	-25.09	-28.08	-25.9	-27.36	-28.3	-27.34	-28.88	-31.97
3	-25.95	-24.92	-26.59	-26.25	-27.49	-27.04	-26.18	-27.58	-29.32	-28.12	-30.97
4	-26.39	-24.8	-26	-25.43	-27.84	-25.78	-25.32	-27.97	-29.19	-28.58	-31.45
5	-27.24	-24.88	-26.96	-24.89	-26.75	-26.43	-25.71	-27.53	-29.04	-27.93	-30.83
6	-26.72	-25.86	-26.81	-26.11	-27.52	-26.39	-26.15	-28.49	-28.93	-27.46	-30.71
7	-26.52	-25.64	-27.52	-25.58	-28.83	-24.62	-29.33	-27.79	-28.9	-27.82	-29.74
8	-26.27	-25.45	-26.64	-26.2	-27.55	-24.28	-25.03	-28.28	-28.8	-28.52	-30.62
9	-26.85	-25.62	-27.43	-25.23	-25.88	-25.18	-25.39	-30.25	-29.48	-28.27	-29.89
10	-26.65	-27.43	-27.28	-26.78	-27.72	-24.48	-25.61	-26.02	-20.55	-28.42	-30.02
11	-27.18	-27.47	-27.48	-27.3	-25.19	-24.81	-25.5	-27.41	-29.66	-26.82	-29.81

APPENDIX B
Monthly Vertical Profiles

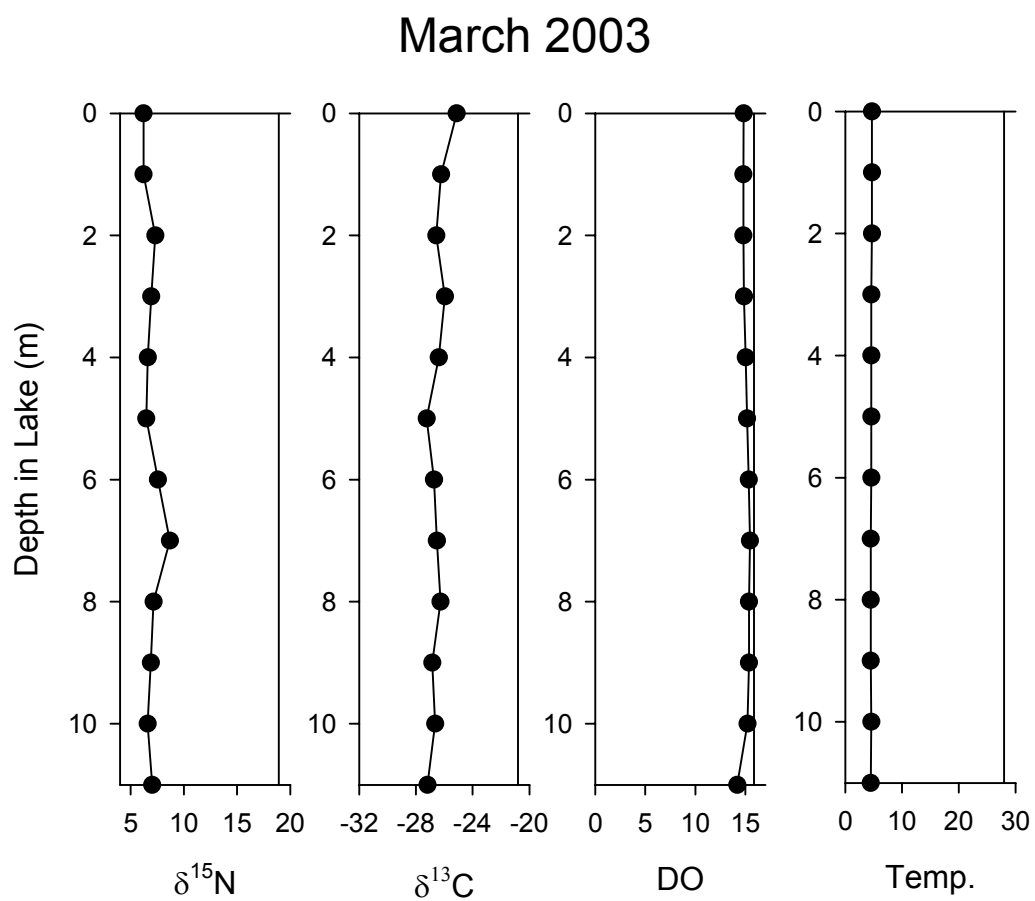


Figure B-1: March 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

April 2003

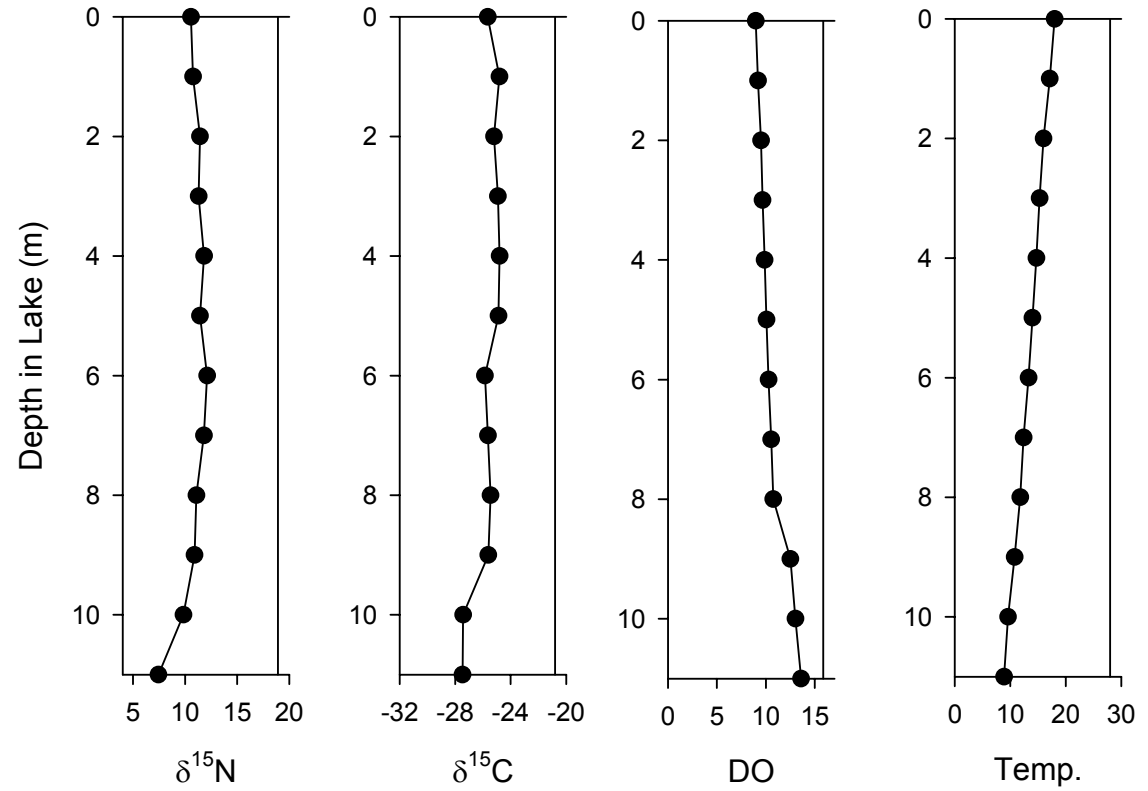


Figure B-2: April 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

May 2003

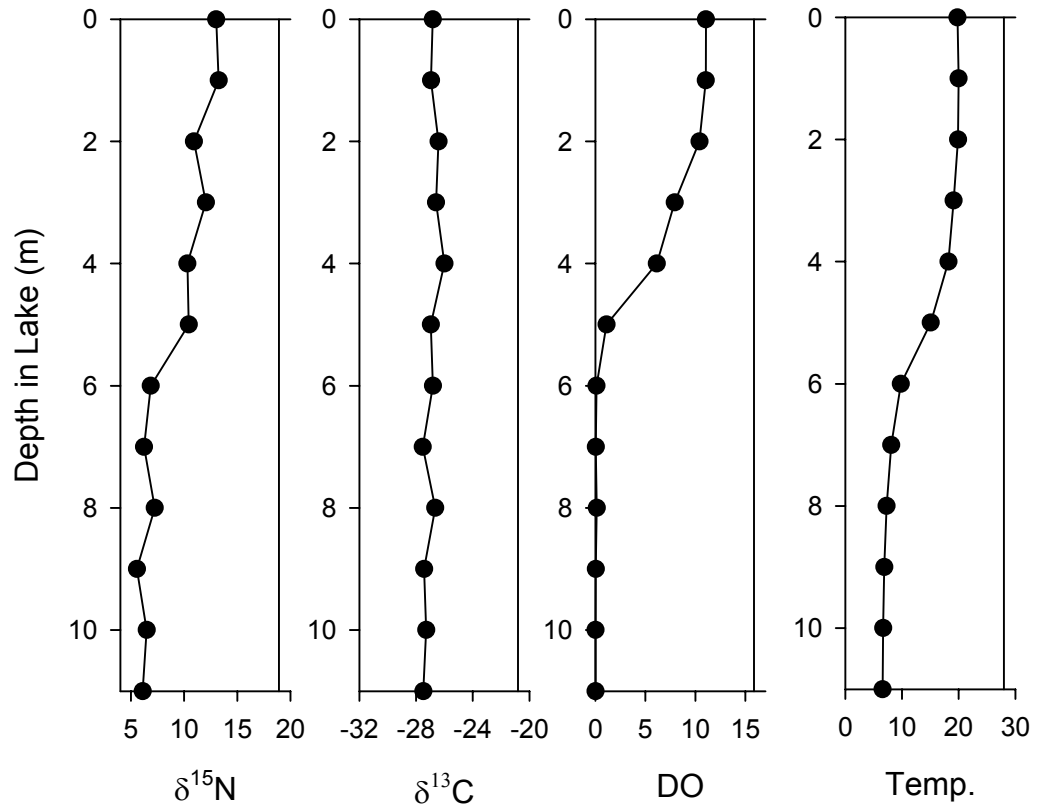


Figure B-3: May 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

June 2003

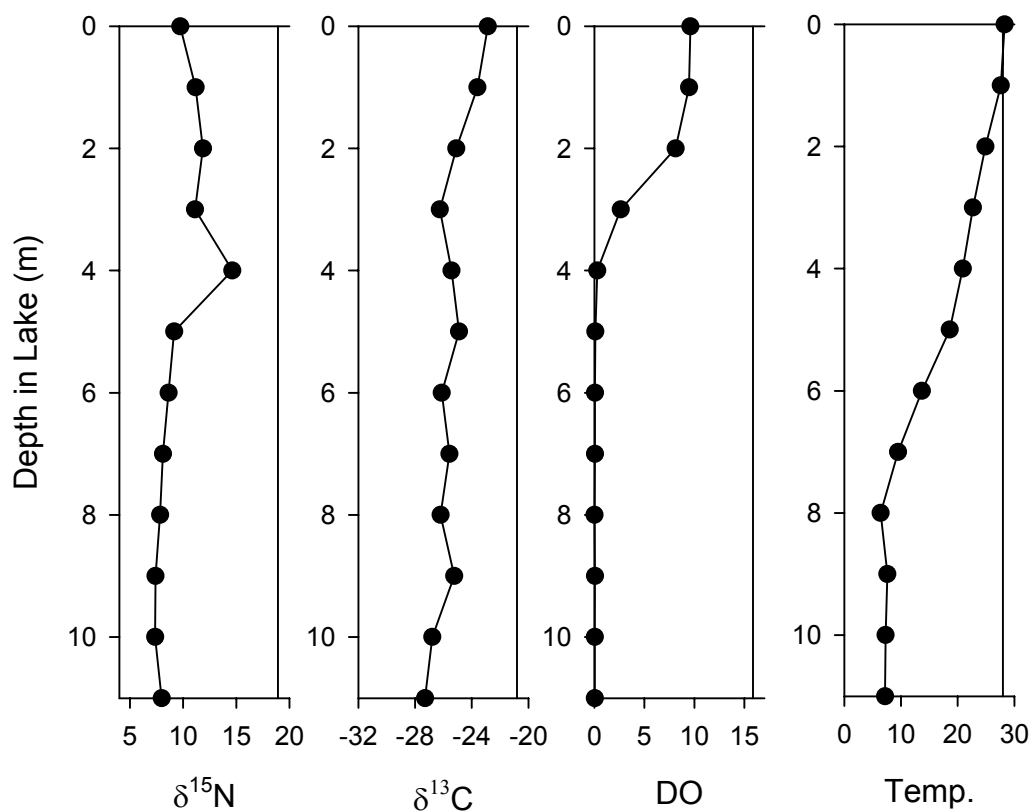


Figure B-4: June 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

July 2003

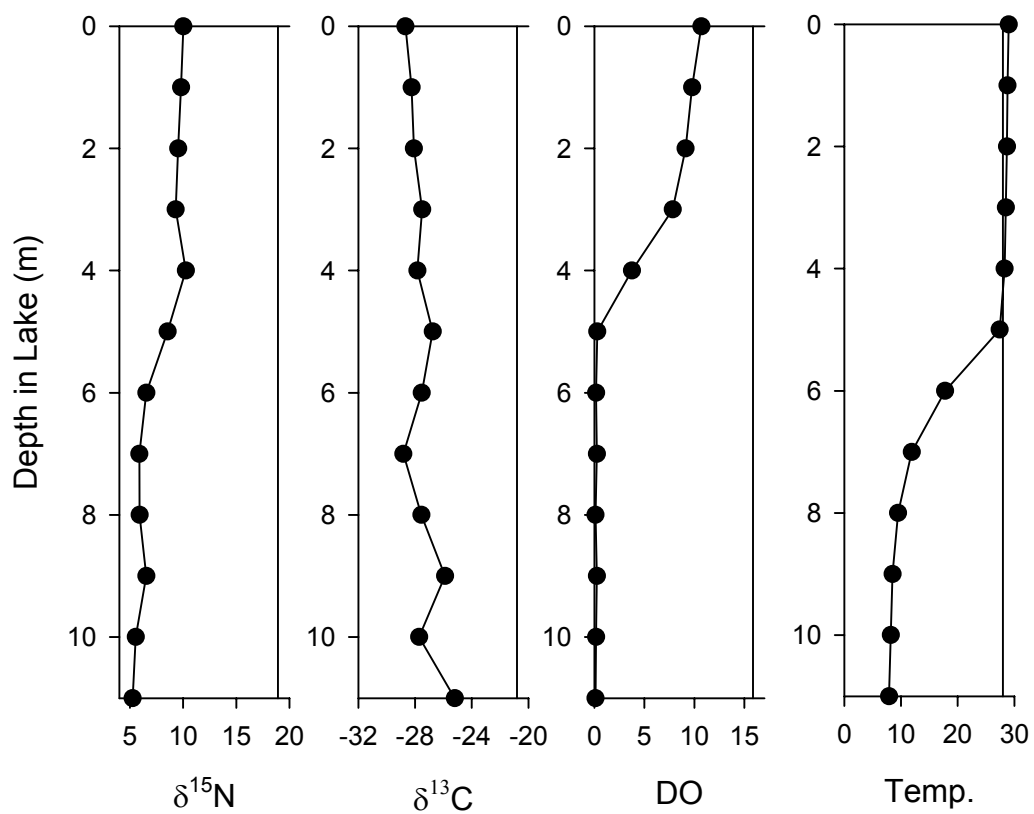


Figure B-5: July 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

August 2003

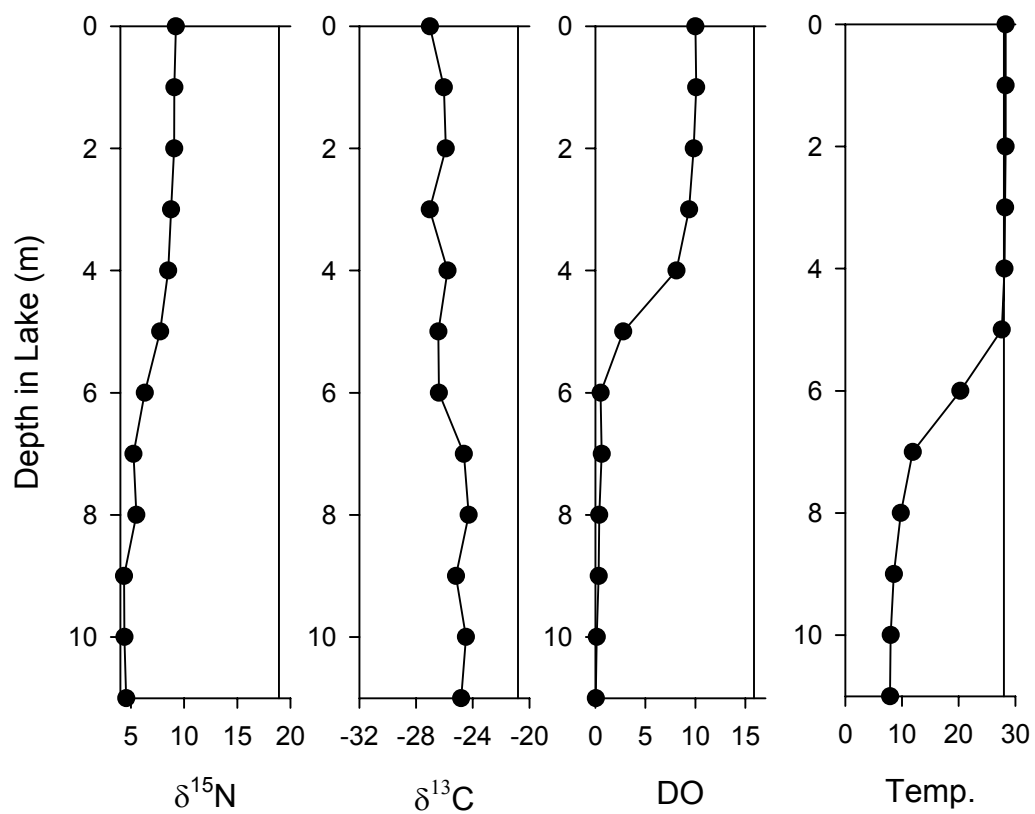


Figure B-6: August 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

September 2003

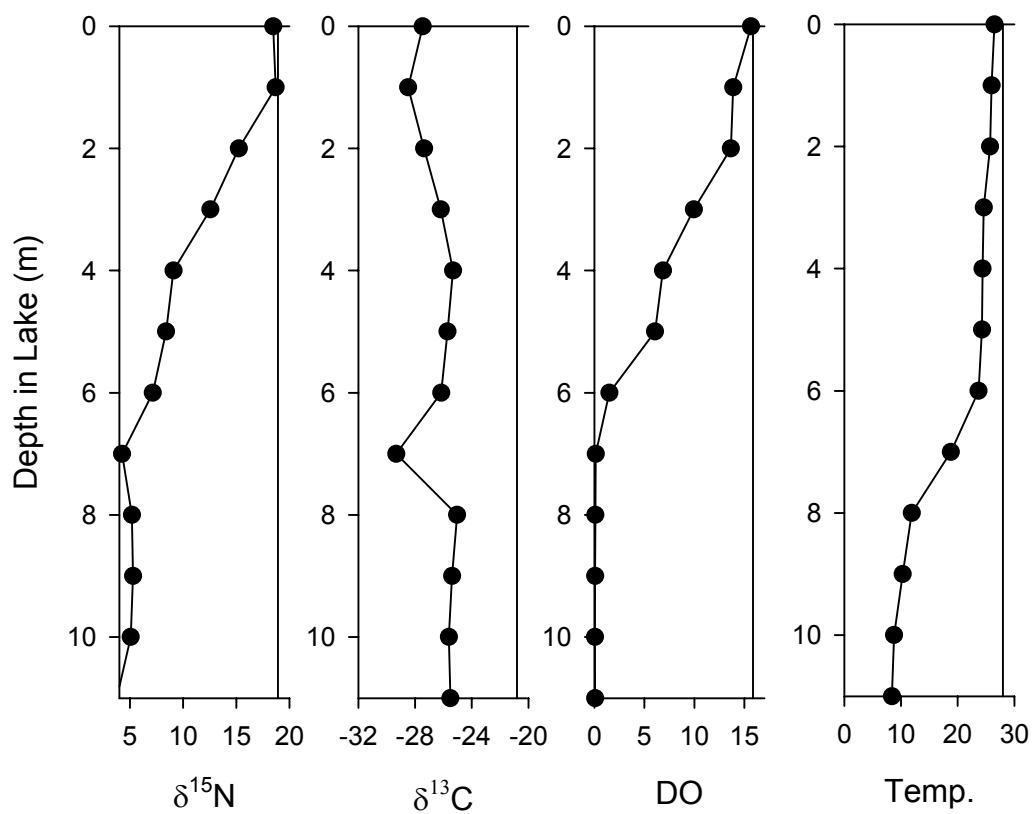


Figure B-7: September 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

October 2003

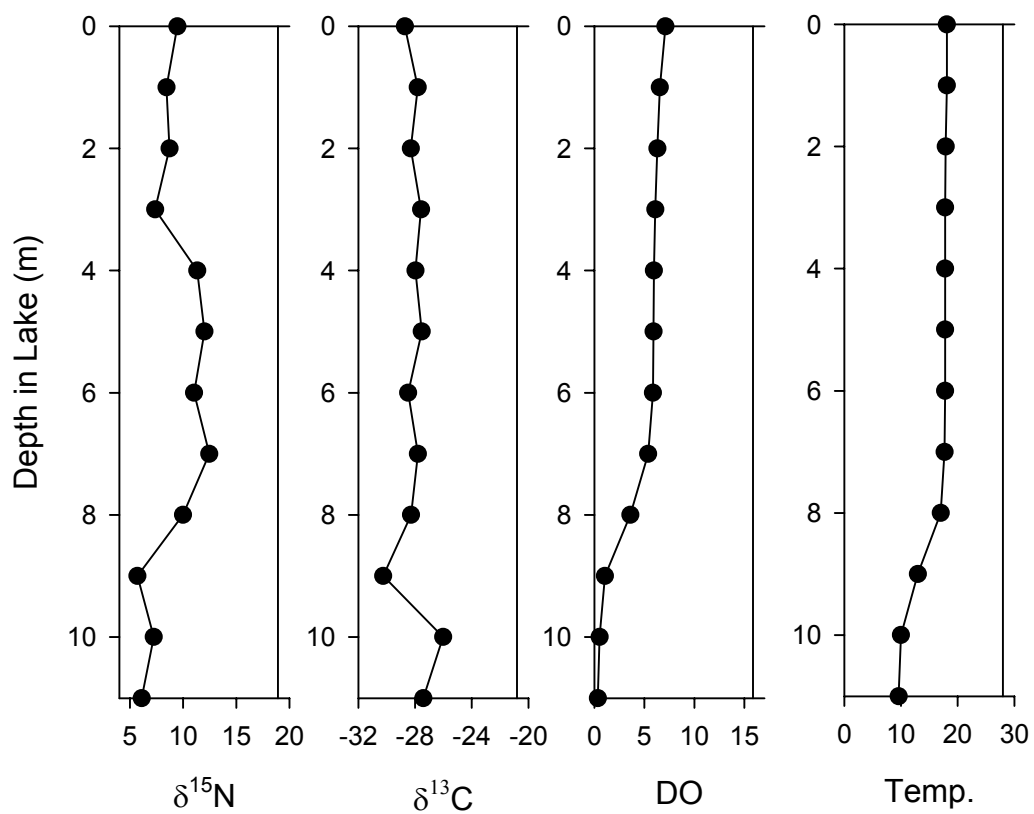


Figure B-8: October 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

November 2003

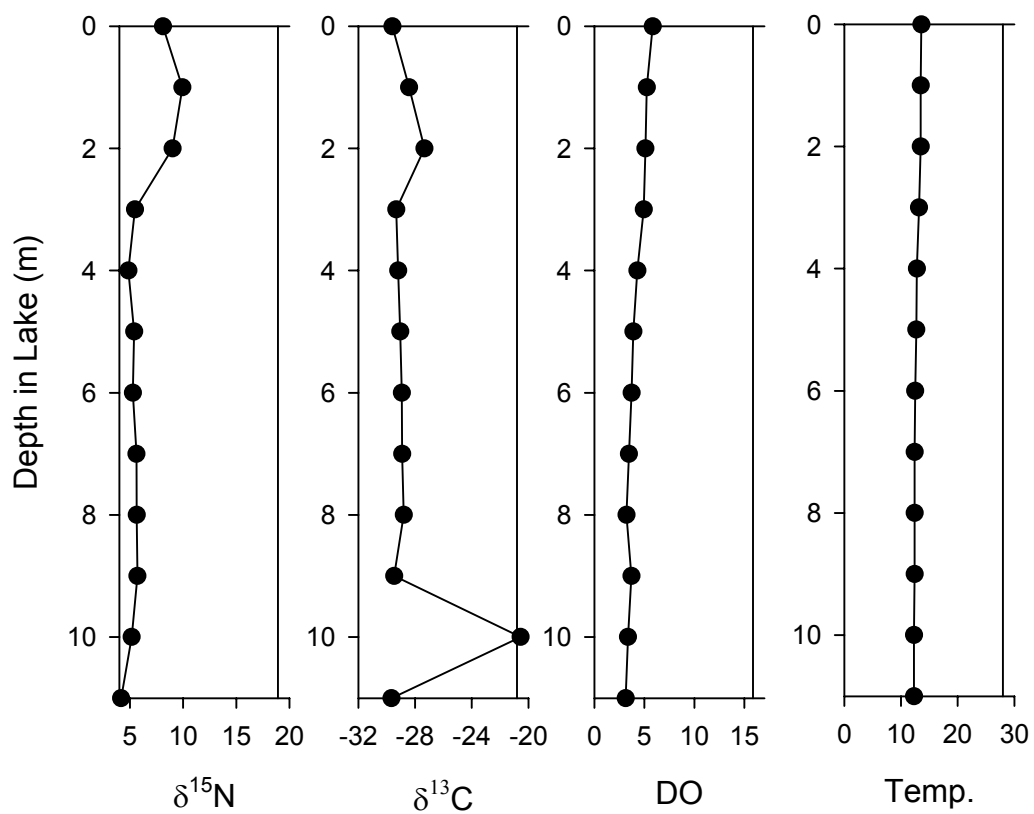


Figure B-9: November 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

December 2003

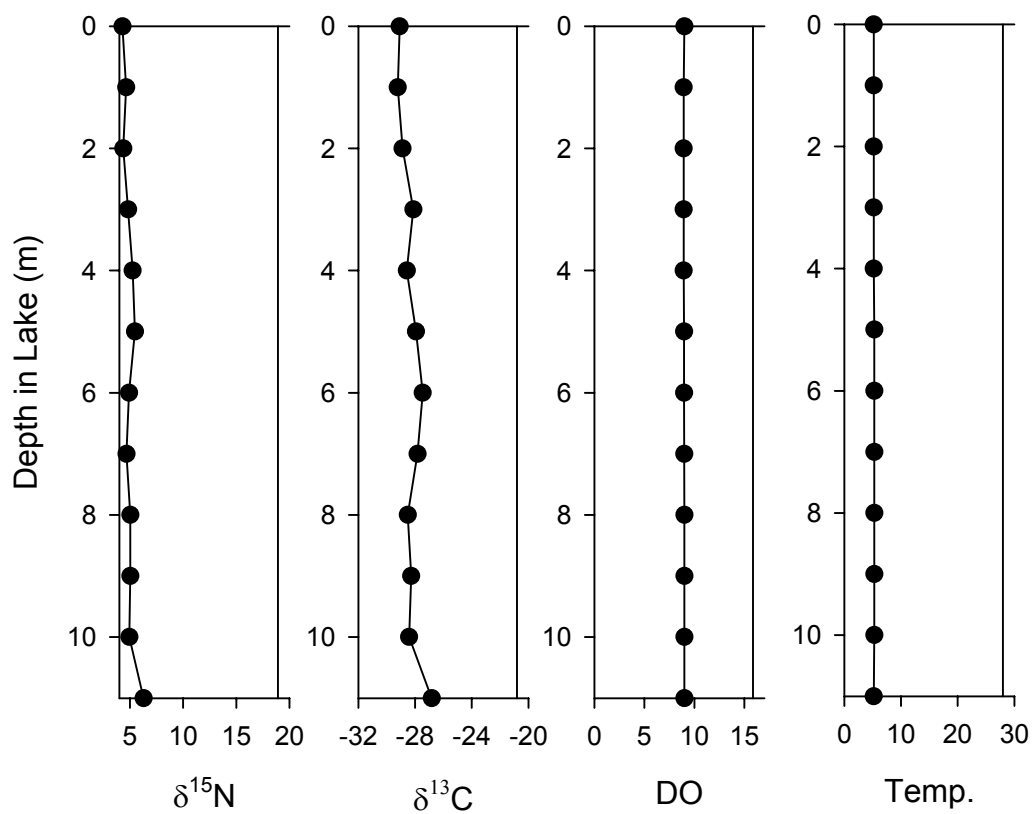


Figure B-10: December 2003 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).

March 2004

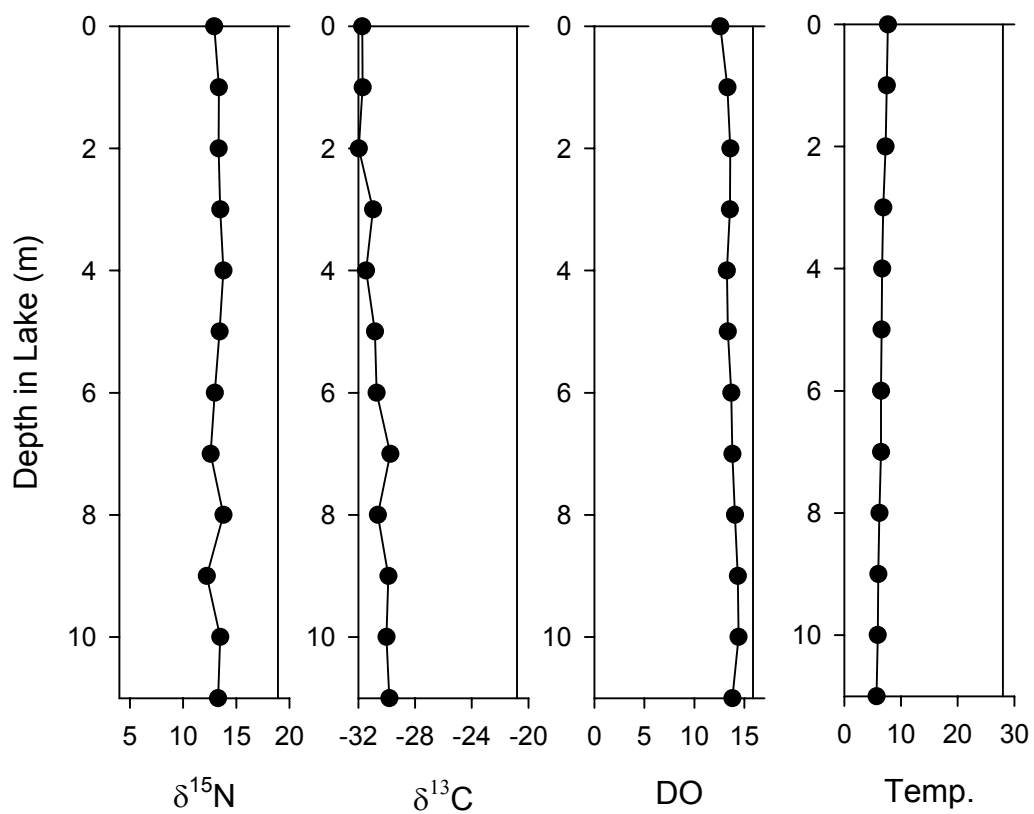


Figure B-11: March 2004 vertical profiles for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of seston, dissolved oxygen (mg/L), and temperature ($^{\circ}\text{C}$).