INFLUENCE OF SPATIOTEMPORAL VARIABLES IN STRUCTURING THE
NEMATODE COMMUNITY OF A FRESHWATER SYSTEM

By

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ABSTRACT

Kyle E. Luth

INFLUENCE OF SPATIOTEMPORAL VARIABLES IN STRUCTURING THE NEMATODE COMMUNITY OF A FRESH WATER SYSTEM

Thesis under the direction of Gerald W. Esch, Charles M. Allen Professor of Biology, Wake Forest University, Department of Biology

The influence of spatiotemporal variables on the structure of the free-living nematode community in a eutrophic lake was studied. In particular, the influence of mo, season, and substratum type was considered. Substratum samples were collected, and nematodes were isolated, identified, and quantified. Prevalence, mean abundance, species richness, and the Shannon-Weiner diversity index (H’) were calculated for (1.) the overall system, (2.) each mo, (3.) each season, and (4.) each substratum type identified in the lake. A total of 2,349 nematodes representing 45 species (morphotypes) from 30 genera were observed, and the overall H’ was 2.5. The most commonly observed genera were Monhystera, Iotonchus, Dorylaimus, Semitobrilus, and Neotobrilus, in that order. Mean abundance for the entire system was 4.9± 0.5 nematodes per samples (mean ± SE). Results suggested that both mo and season influence nematode prevalence, and mo, season, and substratum type all impact the mean abundance of nematodes. Specifically, significant relationships were observed between nematode prevalence and
both month and season. Further, overall nematode mean abundance was significantly higher
in April than other months, lower in winter and higher in autumn than other seasons, and
higher in mixed substratum than any other substrata. Finally, both species richness and
diversity were highest in April, both were highest in spring, and the greatest species
richness was observed in leaf substratum while the greatest diversity was observed in
mud/algae substratum.

The influence of the spatiotemporal factors, season and substratum type, on
nematodes classified by feeding type was also considered. A total of 1,918 free-living
nematodes were classified into 5 feeding types based on the type of food primarily
consumed by each genus. When specific genus-level feeding habits were unknown, the
dominant feeding type observed at the family-level was used. Five feeding types were
observed in the current system: (1.) herbivores, (2.) bacterivores, (3.) unicellular
eukaryote feeders, (4.) predators of animals, and (5.) omnivores. Specifically, 51.8% of
the total nematodes recovered were classified as bacterivores, 21.3% were predators,
14.2% were omnivores, 11.3% were unicellular eukaryote feeders, and 1.3% were
herbivores. Both season and substratum type appeared to influence the community
composition in this system, with season showing the greatest influence on the predator
feeding type, and substratum type showing the greatest influence on both predators and
omnivores. Bacterivores were the most prevalent and the most abundant nematodes in
nearly every season and substratum type considered.

Finally, a second analysis was performed to investigate the influence of
spatiotemporal factors (season and substratum type) on nematodes; instead of classifying
nematodes by feeding type, a second method was used which groups worms by feeding
method. A total of 1,928 free-living nematodes were classified into 5 groups based on the method utilized by each to acquire their food. Five feeding methods were observed in the current system: (1.) deposit feeder/swallower, (2.) epistrate feeder, (3.) chewer, (4.) suction feeder, and (5.) parasitic. In summary, 44.8% of the total nematodes recovered were classified as deposit feeders/swallowers, 34.6% were chewers, 15.6% were suction feeders, 6.4% were epistrate feeders, and 0.4% were parasites. Both season and substratum type appeared to influence the community composition of nematodes utilizing these various feeding methods in the system. Season had the greatest influence on the prevalence of deposit feeders and suction feeders, and on the mean abundance of all feeding methods, with the exception of deposit feeders. Substratum type had the greatest impact on the prevalence and mean abundance of suction feeders and chewers. Deposit feeders were dominant in nearly every season and every substratum type, and showed little fluctuation in both prevalence and mean abundance based on the spatiotemporal factors considered.
CHAPTER I

INTRODUCTION

Nematodes represent one of the most abundant, diverse, and speciose phyla on the planet. They occupy all sediment and substratum types, are found in all climatic zones, and, as a result, are of great ecological importance (Giere, 1993). In most ecosystems, the nematofauna is quite abundant, and freshwater habitats are no exception. Their ubiquitous distribution, high diversity, and short generation time make them ideal organisms for gaining insights into the influence of spatiotemporal factors on the observed patterns of distribution and abundance in a habitat (Bert et al. 2007). This diversity is well exemplified by several studies: Traunspurger (1996a,b) reported 36 freshwater nematode families and 120 species from Königssee, an oligotrophic lake in Germany; Esser and Buckingham (1987) compiled a list of 133 genera containing 160 species of nematodes from freshwaters in North America; and Andrássy (1978) listed 170 genera composed of 605 species from the inland waters of Europe. Furthermore, Andrássy (1992) reported that the number of valid species of free-living nematodes described so far is approximately 11,000. However, even with these large scale estimations, it remains difficult to compare measures of nematode abundance, biomass, or diversity from one freshwater system to the next due to disparity in studies pertaining to the frequency of sampling, the collection techniques used, and the extraction methods employed (Traunspurger et al. 2006).

Historically, the importance of this phylum in structuring and maintaining freshwater ecosystems has been overlooked. Over the course of the past 3 decades, the
integral roles that these important benthic invertebrates play in their environment have begun to gain the attention of researchers. For example, studies have shown that freshwater nematodes are capable of aiding in aquatic plant growth (Ingham et al. 1985), impacting local bacterial community structure (Traunspurger et al. 1997, De Mesel et al. 2004), and are effective for monitoring both quantitative and qualitative aspects of long-term river pollution as bioindicators (Wu et al., in press). Clearly, these organisms play crucial roles in their environments. Accordingly, a better understanding of the factors that contribute to their prevalence, abundance, richness, diversity, and overall community composition would be beneficial. Therefore, the purpose of this research effort was to determine the influence of spatiotemporal factors in structuring the free-living nematode community observed in a local body of water, Mallard Lake, situated in nearby Tanglewood Park.

The following chapter introduces the study site and provides pertinent background on the system utilized in this study. In particular, it includes a brief description of seasonality, predominant substrata, resident and visiting fauna, and other benthic invertebrates observed in the system.

Chapter III reports on the role of spatiotemporal variables, specifically, mo, season, and substratum type, in structuring the free-living nematode community observed. In addition to prevalence and mean abundance for each of these variables, species richness and diversity (based on the Shannon-Wiener diversity index) were calculated using the software package EstimateS (Colwell 2009). Species accumulation curves were also generated by mo, season, and substratum type to better determine the adequacy of
sampling effort and to provide additional support for or against the estimated species richness value observed for each sample.

Chapter IV details the influence of spatiotemporal variables on the community composition of free-living nematodes classified by their feeding preference, i.e., the type of food they primarily consume. This classification system is based on a paper by Yeates et al. (1993), which summarizes the primary food source of soil dwelling nematodes based on genus. Five feeding types (herbivores, bacterivores, unicellular eukaryote feeders, predators of animals, and omnivores) were observed in the current system, and the influences of season and substratum type are discussed in this chapter.

Chapter V describes the influence of spatiotemporal variables on the community composition of free-living nematodes classified by their primary feeding method. In particular, this system classifies nematodes into four groups (deposit feeders/swallowers, chewers, epistrate feeders, and suction feeders) based on buccal morphology described by Traunspurger (1997), and operates under the assumption that one can predict the type of food a nematode typically consumes based on the shape and armament of the buccal cavity. For the purposes of the current study, a fifth feeding method (parasitism) was added to this feeding classification scheme. As in Chapter IV, the influences of both season and substratum type are explored.

Chapter VI is both a summary of the system, and a description of the influence of temporal, trophic, and spatial factors considered in the study. The chapter attempts to tie up the loose ends of the study, while at the same time serves as a cautionary note to those attempting a community-level study aimed at answering the same kinds of questions with regards to the influence of spatiotemporal variables. It provides some insight into
potential pitfalls with this type of study, and includes a brief discussion of the importance of scale in such studies.

Finally, some important notes pertaining to the presentation and discussion of taxonomic groups in this study, as well as comments pertaining to feeding group classifications utilized hereafter need to be addressed. (1.) Unless otherwise specified, all individuals were classified to the level of genus, and not species. Generic names are thus used in the text; however, because individuals were not identified to species, each genus may be represented by one, or more, species. The number of morphotypes contributing to each genus is known and, in the context of the current study, each different morphotype was considered a unique species. Thus, when estimates of species richness or the number of species contributing to a particular genus are provided, they are based on the number of morphotypes involved rather than the number of keyed species. (2.) The two feeding classification schemes were utilized independently, so classification of a nematode based on one scheme in no way influenced its classification based on the other. Taxonomic information alone was utilized for the Yeates et al. (1993) classification system (genus or family), while exclusively buccal morphology was used for the Traunspurger (1997) classification system.
LITERATURE CITED


Traunspurger W 1996a. Distribution of benthic nematodes in the littoral of an


CHAPTER II

STUDY SITE

Mallard Lake is a 4.9 ha public access pond located in Forsyth County, in the Piedmont region of North Carolina (Fig. II-1). The lake is about 26 km SW of Winston-Salem, NC, and is situated in a public-access, county-owned park (Tanglewood Park). It is frequented by fishermen and non-motorized watercraft (predominantly during the summer mo). The NE end of the lake is bordered by a golf course, and the NW and W banks by continuous woods. Throughout the study (January-October, 2009), the lake substratum temperatures reflected all four seasons, ranging from lows of 4 C in January to highs of 34 C in May and July. During the winter mo, ice formed along the perimeter of the lake, especially along the W bank. Due to the shade of the trees, this region tended to experience cooler temperatures than other sites around the pond on a year-round basis. The pond has been described as eutrophic and is believed to experience frequent turnover (Esch, pers. comm.). Multiple substrata are present in the pond and the most prevalent include leaf litter (organic matter, stick, and other detritus), mixed (equal parts leaf cover and exposed mud/clay substratum), mud/clay, and mud/algae. A defined littoral zone is present around most of the perimeter of the lake, with the exception being a 400 m stretch along its W bank. In this area, there is a substantial amount of leaf litter present, and solid clay substratum is often absent, even at water depths of a m, or more. Surveys of the pond conducted over the course of the present study determined that it is habitat for a number of fish, birds, reptiles, and amphibians, and is frequented by various mammals, including raccoons and opossums. Additionally, a robust and diverse community of
benthic invertebrates is present in the lake. The meio- and macrobenthos include four species of snail, ostracods, copepods, oligochaete worms, multiple species of insect larvae, a host of parasitic flatworms and roundworms, and a large number of free-living nematodes.
Fig. II-1: Map of Mallard Lake.
CHAPTER III

The Influence of Spatiotemporal Factors on the Community Composition of Free-living Nematodes in a Freshwater System

ABSTRACT

Few studies have attempted to better understand the influence of abiotic and biotic factors on the composition of the free-living nematode communities in freshwater systems. The current study was undertaken to discern the role of spatiotemporal factors, i.e., mo, season, and substratum type, on the community composition of these benthic invertebrates in a eutrophic lake. Substratum samples were collected from January to October, 2009, from Mallard Lake. Nematodes were the second most prevalent benthic invertebrate observed (ostracods were first) and were recovered from 65.7% of the samples collected. A total of 2,349 nematodes was recovered with a mean abundance of 4.9 ± 0.5 per sample, and a mean density of 4.9 ± 0.5 per 55 cm³ (mean ± SE). A total of 45 species from 30 genera was identified, and the Shannon-Wiener diversity index for the entire system was H’=2.5. Monhystera was the dominant nematode genus, although species of Iotonchus and Dorylaimus were also observed in large numbers. Both prevalence and mean abundance varied greatly, collectively suggesting that season plays an important role in determining the presence and number of nematodes in this system. Mean abundance was significantly lower in winter and significantly higher in autumn than other seasons. There appeared to be no influence of substratum type on prevalence; however, mean abundance was significantly higher in mixed substratum than other
substrata. April had both the greatest species richness and the greatest Shannon diversity. Spring also exhibited both the greatest species richness and the greatest Shannon diversity. Leaf substratum possessed the greatest species richness, while mud/algae substratum had the greatest Shannon diversity. Mo, season, and substratum type all influenced the community composition of the nematofauna in this system.
INTRODUCTION

Relatively little work has been done to discern the impact of spatiotemporal factors on the structure of free-living freshwater nematode communities. Over the past several decades, more studies have begun to elucidate the integral contributions of these organisms to the success and longevity of the freshwater ecosystems they inhabit. For instance, free-living aquatic nematodes are both capable of aiding in plant growth (Ingham et al. 1985), and helping to structure local bacterial communities (Traunspurger et al. 1997, De Mesel et al. 2004). In soil ecosystems, nematodes play important roles in nutrient recycling by liberating minerals from the plant tissues and microorganisms they consume, thereby facilitating absorption by nearby plant roots (Gomes et al. 2003). Furthermore, free-living, aquatic nematodes can be readily employed as bioindicators for monitoring both quantitative and qualitative aspects of long-term environmental stressors, e.g., river pollution (Wu et al., in press). Such studies make it clear that understanding the impact of these benthic invertebrates on one another, as well as on the other organisms with which they share their habitat, is critically important. Furthermore, understanding how spatiotemporal factors influence the observed community structure of the free-living aquatic nematodes in this system is also of particular interest.

In the few studies that exist looking at the influence of temporal variables on the free-living nematode community structure in freshwater systems, it is clear that these factors do, indeed, play an important role. Vidaković and Bogut (2004) reported that the abundance and diversity of nematodes in Sakadaš Lake (Croatia) was correlated to seasonal temperature variations. Similarly, Traunspurger (1996a,b) reported that seasonality influenced observed nematode abundances in Lake Königssee (Germany), as
did Nalepa and Quigley (1983) in Lake Michigan (United States). These findings make it clear that further understanding of the influence of temporal factors on the distribution and abundance of free-living nematodes is vital to fully understanding the processes that are driving the observed patterns of nematode assemblage in this freshwater system.

Studies conducted in marine habitats have attempted to understand the role of environmental processes on the spatial distribution of free-living nematodes and suggest that meiofauna (nematodes included) is commonly dispersed via passive transport through the water column (Hagerman and Rieger 1981, Palmer 1988, Atilla and Fleeger 2000). One of the driving forces behind this type of dispersion is the substantial changes in habitat structure that occur in lake littoral zones due to periodic disturbances, e.g., wind events, wave action, or fluctuations in water level (Peters et al. 2007b). A study by Traunspurger and Drews (1996) showed that the free-living nematodes inhabiting the littoral zone of a lake occurred in the greatest proportions in the top 2 cm of substratum, and several studies have suggested that the meiobenthos near the water-substrate interface are more susceptible to erosion (Warwick and Gee 1984, Eskin and Palmer 1985, Palmer and Gust 1985). However, the free-living nematode assemblage is not entirely dependent on such random dispersion events, because these benthic invertebrates are actually capable of habitat selection.

Ullberg and Ólafsson (2003) reported that nematodes also possess the ability to actively choose preferred habitat. Furthermore, Chandler and Fleeger (1983) reported that in a marine system, nematode dispersal occurred equally well via passive, water-column transport and active horizontal migration. Thus, nematodes are capable of choosing the areas they inhabit, a characteristic that allows them to rapidly colonize
substrates in both marine (Sherman and Coull 1980) and freshwater systems (Peters et al. 2007a). Furthermore, Pehofer (1989) and Traunspurger (1996a) both reported that abiotic parameters of habitat, e.g., sediment type, water depth, water temperature, concentration of dissolved oxygen, and quantity of available food, have a significant influence on the distribution and abundance of nematodes in both marine and freshwater systems, respectively. Understanding which habitats nematodes prefer following passive or active dispersal in the water column, or active colonization following a disturbance event is of great interest. A better understanding of how free-living nematode selectivity of substratum types influences their observed community structure would provide a first step in understanding their role in affecting the community structure of other, closely associated, organisms in the system.

The purpose of the present study was to determine the influence of spatiotemporal factors on the community composition of the free-living nematofauna in this system. Nematode prevalence and mean abundance were used as measures of the affect of mo, season, and substratum type on the community structure of these organisms. Nematodes were recovered from substratum samples via Baermann extraction techniques, isolated, and identified to genus. Species richness and the Shannon-Wiener index for diversity were calculated for: (1.) the entire system, (2.) each mo, (3.) each season, and (4.) for each substratum type observed. Species accumulation curves were generated for each mo, season, and substratum type to evaluate the appropriateness of each species richness estimate in all of these samples.
MATERIALS AND METHODS

Sampling method

Samples were collected from Mallard Lake, a 4.9-ha eutrophic lake located in the Piedmont region of North Carolina. Its perimeter measured 1,480 m and was broken into 148 equal transects measuring 10 m each. Each transect was assigned a number (1-148) and was classified as leaf, mixed (containing nearly equal amounts of leaf and mud/clay), mud/algae, or mud/clay substratum based on what was most frequently observed at 2.5, 5, and 7.5 m intervals along each transect. Those containing multiple substrata were ultimately narrowed down to a single substratum type based on the littoral zone contents observed most frequently therein. For each sampling date, a random number generator was used to select three transects from each substratum type. To decrease the amount of site specific re-sampling, each transect was further reduced into 40, 0.25 m subsections, and again a random number generator was used to select two sites within each transect from which samples were taken. This ensured that 24 independent samples were obtained from 12 transects during each collection.

All samples were collected at a distance of about 1.5 m from the shoreline where the water level was typically around 1 m. Water depth was much more variable in transects containing predominantly leaf substratum, especially those along the western bank of the lake where a defined littoral zone was often absent. Samples were collected by filling 70-ml (ID 4.7 cm, 4.6-cm length) screw cap containers with substratum. Containers were transported to the lab and the contents were inspected for nematodes.

Mo were grouped into seasons based on naturally occurring breaks in substratum temperature (Appendix A-1 & A-2). Winter was from January 29 through March 26,
spring was from April 2 through May 8, summer was from May 28 through August 18, and autumn was from September 10 through October 28, with all sampling occurring in 2009.

**Nematode recovery**

Typically, 55 ml of each sample was transferred from its respective container to an individual Baermann-funnel apparatus; however, depending on the substratum being analyzed, this volume varied slightly (about ±5 ml). The Baermann technique was employed because of its ease of use and its utility for extracting nematodes from a wide range of substratum types (Hodda and Eyualem-Abebe, 2006). Baermann extractions were run for 18-24 hr, and all nematodes, benthic invertebrates, and substratum that settled to the bottom of each apparatus were transferred into separate 15-ml centrifuge tubes. Tubes were filled to a total volume of 15 ml with aged tap water and centrifuged at about 700 rpm for 2.5 min. About 11 ml of supernatant was removed, and the remaining sample and water were transferred to a 51-mm petri dish. Each dish was observed independently using a stereomicroscope, and all nematodes were isolated on glass slides for further observation. Nematodes were immobilized by adding 95% ethanol or by heat killing, and were quantified throughout the course of the study.

Identification of nematodes to morphotype (using both dry and oil immersion high-powered objectives on a bright field microscope) began with the March 29, 2009 collection, and continued throughout the remainder of the study. Representatives of each morphotype were drawn or photographed using a microscope-mounted Canon Rebel T1i digital camera. When possible, individuals were transferred into 95% ethanol or glutaraldehyde for future sequencing or observation via SEM, respectively.
The presence or absence of other benthic invertebrates was noted beginning with the samples collected on June 22, 2009, and continued throughout the remainder of the study, i.e., the final 208 samples collected. With the exception of nematodes, benthic invertebrates were not quantified.

Data analysis

A total of 2,349 nematodes was enumerated in the current study, and all individuals were utilized in the statistical analyses reported in this chapter, regardless of taxonomic or feeding type classification.

Presence/absence data were utilized to determine whether the number of sites containing nematodes differed based on mo, season, and substratum type. \( \chi^2 \)-tests were conducted comparing the observed presence values for mo, season, and substratum type to expected presence values based on chance alone. Expected values were generated by averaging the observed presence values for each set of comparisons. A correction factor was applied to expected values to account for differences in sample size. Prevalence values were also calculated using observed presence data for each data class (mo, season, or substratum type).

Data collected for the current study consistently failed the \( F_{\text{max}} \) test for equal variance. To decrease the chance of committing type II (false negative) errors, these data were not analyzed with standard parametric statistical tests, e.g., ANOVA. Ordinarily, when the assumptions of normality and equal variance are not satisfied, a non-parametric analog, e.g., the Kruskal-Wallis test, can be employed. However, the use of these tests incur limitations in their own right, including most notably, a loss of statistical power when compared to parametric tests. Therefore, the current study utilized Monte Carlo
randomizations to determine the significance of differences between means within and among samples.

This method of analysis offers a powerful alternative to parametric tests because it randomly re-samples the collected data set, thereby removing the constraints of normality and equal variance encountered when using parametric tests. Specifically, the difference in mean abundance values for every possible pairwise combination of mo, season, or substratum type was calculated. The data set was then randomly rearranged 30,000 times, and the pairwise differences in mean abundance between each mo, season, or substratum type were re-calculated from the randomized data set. The calculated pairwise differences resulting from the 30,000 random iterations were then compared to the actual differences in mean abundance observed between mo, seasons, or substratum type in the system, i.e., the actual observed mean abundance differences, prior to random rearrangement. P-values were generated by dividing the number of times the randomized mean difference was equal to or exceeded the actual observed mean difference between mo, seasons, or substrata, by the total number of iterations. When the iterative mean abundance differences were greater than or equal to the actual mean abundance differences more than 5% of the time, the differences in actual observed means were not considered significant.

Regression analysis was utilized to determine whether a significant relationship existed between prevalence and season. Regression analysis was also performed to determine whether a significant relationship existed between mean abundance and season. Spearman’s ρ was used to determine whether species richness and diversity were correlated based on mo, season, and substratum type. Additionally, Spearman’s ρ was
used to determine whether species richness and diversity were correlated with substratum temperature based on both mo and season.

Of the 2,349 nematodes collected, only 1,828 were identified to morphotype. This discrepancy was the result of one of three factors: (1.) morphotyping did not begin until the 26 March 2009 collection, so the identity of any nematodes collected prior to this date is unknown; (2.) some nematodes were too small to be clearly identified; or (3.) a very small number of nematodes were lost during transfer from the stereomicroscope to the bright field microscope. Species richness (S) was used as a count of the total number of species observed in the whole system, as well as in each mo, season, and substratum type. This measure was utilized due to its sensitivity to rare species, i.e., rare species are still counted. The Shannon-Wiener diversity index (H’) was used as a measure of diversity and evenness in the current study. This index is based on the proportional abundance of species in a sample and maintains moderate sensitivity to both rare species, and common species. Dominance indices, e.g., Simpson’s index (D), are also based on the proportional abundance of species; however, they are more weighted toward the common species (Magurran 1988), and tend to ignore rarer species. For this reason, the Simpson’s index was not utilized in the current study. The software package EstimateS (Colwell 2009) was used to calculate the Shannon index, and the Sobs (Mao Tau) values utilized for creating the species accumulation curves. Species accumulation curves were generated as a means of determining the validity of the reported species richness estimates for each mo, season, and substratum type. The significance threshold for all statistical tests utilized in this study was set at \( p \leq 0.05 \).
**Terminology**

Presence was defined as the total number of samples containing 1, or more, nematodes. The use of the terms prevalence and mean abundance was adapted from Bush et al. (1997) to fit the context of the current study. The term prevalence was defined as the number of sites containing 1, or more, nematodes, i.e., presence, divided by the total number of sites observed, and was expressed as a percent. The term presence was used to describe sites containing 1, or more, nematodes. Mean abundance was defined as the average number of nematodes per sample, where all sampling units (sites) were included, regardless of whether or not they contained nematodes. In the current study, a sampling unit was equal to 55 cm$^3$. Thus, a mean abundance of 3 nematodes can be equally expressed as either 3 nematodes/sampling unit or 3 individuals/55 cm$^3$ (the latter representing a measure of mean density). All mean abundance values were reported as the mean (± SE).

Unless otherwise specified, all individuals were classified to the level of genus, and not species. Generic names are thus used in the text; however, because individuals were not identified to species, each genus may be represented by 1, or more, species. The number of morphotypes contributing to each genus is known and, in the context of the current study, each different morphotype was considered a unique species. Thus, when estimates of species richness or the number of species contributing to a particular genus are provided, they are based on the number of morphotypes involved rather than the number of keyed species.
RESULTS

Nematodes versus other benthic invertebrates

A total of 2,349 nematodes was observed in 480 independent substratum samples. Nematodes were present in 65.7% of the total samples observed. In the final 208 samples collected, i.e., those in which the presence or absence of other benthic invertebrates was documented, nematodes occurred with a prevalence of 82.3%. Ostracods were observed in nearly the same proportion of samples as free-living nematodes (83.6%). They were the only benthic invertebrate to exceed the prevalence of nematodes in this system. Other commonly observed benthic invertebrates included rotifers (49.0%), *Atrichopogon* spp. (midge) (37.0%), various flatworm species (29.3%), *Bezzia* spp. (midge) (24.5%), oligochaetes (19.7%), and copepods (15.9%). Additionally, diatoms were observed in 53.4% of samples collected.

Influence of mo, season, and substratum type on nematode prevalence

Overall nematode prevalence for the entire system was 65.7% (Table III-1). Figure III-1 shows that monthly prevalence followed a definite bimodal pattern, peaking in January at 75.0%, dropping to a constant trough from February to June (ranging from 39.6-55.9%), and increasing to a constant peak from July to October (ranging from 79.2-91.7%). The observed monthly presence values differed significantly from expected based on the assumption that all samples collected were equally likely to contain nematodes ($\chi^2$ test, p<0.001). The greatest deviations from expected presence values occurred in the final four mo of the study, and, in all cases, the observed presence (number of sites containing nematodes) was greater than expected by chance. No
significant relationship existed between prevalence and mo over the course of the study ($R^2=0.358$, $p=0.067$).

Prevalence showed a steady increased from one season to the next, ranging from 48.8% in winter to 84.4% in autumn (Table III-1, Fig. III-3), and the relationship between prevalence and season was significant ($R^2=0.964$, $p=0.0184$). In addition, observed presence between seasons differed significantly from what was expected ($\chi^2$ test, $p<0.001$), assuming all samples in all seasons had equal chances of containing nematodes. The greatest deviations from expected values were observed in winter, spring, and summer. In the latter, presence was greater than expected, while in winter and spring presence was less than expected.

Prevalence values were very similar in leaf, mud/algae, and mud/clay substrata, and were consistently lower than the nematode prevalence observed in mixed substratum (Table III-1, Fig. III-5). Differences between observed and expected presence values were significant ($\chi^2$ test, $p=0.016$). The greatest deviations from expected values were observed for leaf, mud/algae, and mud/clay substrata. Nematodes were recovered from fewer sites than expected in the former substratum type, while more sites than expected contained nematodes in the mud/algae and mud/clay substrata.

**Influence of mo, season, and substratum type on the mean abundance of nematodes**

Nematode abundance was highly variable across mo, but followed, with the exception of April, the same general trend as prevalence (Fig. III-2). During April, prevalence was low (55.9%), while mean abundance was high ($5.7 \pm 1.3$), relative to other mo (Table III-1). Overall, mean abundance reached a minimum in June and peaked in July (Fig. III-2). Mean abundance in April and July was significantly higher than in
surrounding mo (see Appendix B-1 for p-values). Mean abundance was significantly lower in January, February, March, May, and June than all other mo, although none of these mo differed significantly from one another.

Similar to the trend observed for seasonal prevalence, mean abundance also exhibited a positive trend over the course of the study (Table III-1, Fig. III-4). Furthermore, the relationship between mean abundance and season was significant ($R^2=0.920$, $p=0.041$). Mean abundance was significantly lowest in winter and significantly highest in autumn (Appendix B-2, Fig. III-4). Although values in spring and summer did not differ significantly, a minimal increase was observed from spring to summer (Table III-1, Fig. III-4).

Finally, mean abundance was significantly higher in mixed substratum than in all other substrata (Table III-1, Fig. III-6); however, differences between leaf, mud/algae, and mud/clay substrata were not significant (Appendix B-3).

**Nematode community composition**

In all, 45 species from 30 genera (based on 1,828 morphotyped individuals) were recovered (Appendix C-1). This finding is in accordance with Traunspurger (2002) who reported that species richness in lakes and rivers tends to be between 30 and 70. The overall Shannon diversity index ($H'$) for the nematode community in the present system was 2.5. The family Monhysteridae was the most abundant (comprising 42% of the total nematode fauna) and the most speciose (with 13 species), i.e., morphotypes, represented. Two species from this family contributed 35.9% of the relative abundance observed. Mononchidae was the second most speciose family observed (3 morphotypes), and the fifth most abundant, contributing only 4.1% of the total nematodes recovered. The
second most abundant family was Anatonchidae, which contributed 17.1% of the total abundance, and was represented by only two species. All other families were less abundant, and less speciose.

Table III-1 also shows the community richness and diversity parameters calculated for each mo, season, and substratum type. Species richness was highest in April, and lowest in March. It is likely that the latter value is underestimated and an artifact of the small sample size in March (morphotyping of nematodes did not begin until the final series of samples in this mo). All other mo show similar values of species richness, ranging from 14 to 19 species. The Shannon index was highest in April and August, and lowest in March and May (Table III-1). The peak in this index in April corresponded with a peak in richness; however, there was not a significant correlation between these two parameters across all mo (Spearman’s $\rho=0.55$, $p>0.05$).

Species richness was highest in the spring and was markedly lowest in autumn. Similarly, nematode diversity ($H'$) peaked in spring, and was lowest in autumn. Overall, the values for the Shannon index tended to be much more similar for all seasons than were species richness values (Table III-1). Species richness was strongly correlated with Shannon diversity based on season (Spearman’s $\rho=1.00$, $p<0.001$).

Finally, substratum type appeared to have little influence on species richness or $H'$ as both series of values were very similar across all substratum types. Leaf substratum showed the highest species richness; however, the highest value for Shannon diversity was not observed in this substratum type. Rather, the greatest $H'$ was observed in mud/algae substratum.
Species richness and Shannon diversity were not correlated based on substratum type (Spearman’s $\rho=-0.80$, $p=0.20$). Both species richness and Shannon diversity, however, were correlated with substratum temperature across seasons (Spearman’s $\rho=0.27$, $p<0.001$ and Spearman’s $\rho=0.39$, $p<0.001$, respectively), but neither was correlated with substratum temperature across mo (Spearman’s $\rho=0.09$, $p=0.10$ and Spearman’s $\rho=0.07$, $p=0.17$, respectively).

Species accumulation curves, based on the number of sites sampled, suggested that the sampling effort in the current study may have underestimated the actual species richness in some mo, seasons, and substrata (Appendix D-1-3). Obvious examples of this underestimation were observed for the curves corresponding to March, May, June, and winter where a plateau was not reached. This suggests that more sites would need to be sampled during these periods to improve the likelihood of actually estimating the true species richness. Even so, the mo species accumulation curves supported the assertion that species richness was much higher in April than any other mo (Appendix D-1).

Similarly, the season species accumulation curves showed a clear ranking of seasons such that spring was the most speciose season, followed by summer, autumn, and finally winter (Appendix D-2). However, the placement of winter in this sequence is unclear due to the small sample size in this season. Resolution of the accumulation curves for substratum type was less clear. It appeared that the most probable ranking from the most species rich to the least was mixed, leaf, mud/clay, and mud/algae substrata (Appendix D-3). The accumulation curve for leaf substratum appeared to be furthest from reaching its asymptote. Thus, the species richness was likely underestimated for this substratum type.
Monhystera was the dominant genus in all mo (ranging in relative abundance from 20.5-71%) except in March, April, and July when Terschellingia, Semitobrilus, and Iotonchus were the dominant genera, respectively. In July, however, individuals of Iotonchus and Monhystera occurred in nearly equal proportions (37.8 and 36.7%, respectively). Similarly, Monhystera was the dominant genus in every season (30.4-44.8%) except in winter (4.2%). During this time, Terschellingia and Dorylaimus dominated, contributing 41.7 and 25% of the total nematodes observed, respectively. During the spring mo, Semitobrilus was the next most abundant, followed by members of Prionchulus and Monochromadora, which occurred in the same proportion. During the summer mo, Iotonchus was the second most abundant genus, followed by Dorylaimus. Finally, during the autumn mo, Iotonchus and Dorylaimus were the second and third most abundant genera, respectively, though they appeared in much more similar proportions during these mo than during the summer mo. Monhystera spp. dominated all substrata (ranging in relative abundance 27.5-43.0%) except mud/algae (34.6%) where the most abundant genus was Iotonchus (38.9%). Semitobrius was the second most dominant genus in leaf substratum, and both Monhystera and Semitobrilus were found in much greater proportions here than were other genera. Dorylaimus was the second most abundant genus in mixed substrata, and occurred greater than two times more frequently than did the third most abundant genus, Iotonchus. The latter dominated the mud/algae substratum type, though individuals of this genus appeared in nearly equal proportions as those of Monhystera. Members of Dorylaimus were notably absent in this substratum type. Monhystera was found more than twice as frequently in mud/clay substratum as were individuals representing the second and third most abundant genera, Iotonchus and
Dorylaimus, respectively, which occurred in nearly equal proportions in this substratum type.
DISCUSSION

Benthic invertebrates

With the exception of ostracods, nematodes were the most prevalent benthic invertebrates observed in this system. Nematodes commonly dominate the meiobenthos in freshwater systems, including both lotic (Anderson 1992, Eisendle 2008) and lentic (Vidaković & Bogut 2004, Bergtold & Traunspurger 2005) habitats. Although the abundance of other macro- and meiobenthos were not quantified in the current system, nematodes did appear to occur in greater numbers than other benthic invertebrates.

The most abundant nematode family was Monhysteridae, accounting for 42% of the total nematodes recovered. Similar dominance by this family of bacterivorous nematode was reported from a eutrophic lake (Michiels and Traunspurger 2004) and from a series of eutrophic farm ponds (Bert et al. 2007). Monhystera was the dominant genus in this system and contributed >35% of the total nematodes observed. This genus has been reported with high abundance in oligotrophic lakes (Strayer 1985), and Wu and Liang (1999) suggest that nematodes of this genus cannot normally thrive in eutrophic systems. The results of the current study suggest otherwise. Furthermore, Prejs (1977) showed that Monhystera was present in both oligotrophic and eutrophic lakes, but that the genus was represented by more species in the former than the latter. The study added that one species (Monhystera paludicola), of those species occurring in lakes of either trophic, was among the most abundant species recovered from either lake type.

Influence of mo and season

Monthly prevalence and monthly mean abundance followed similar patterns over the course of the current study (Fig. III-1 & III-2, respectively). Prevalence peaked in
July, and was lowest in February. Mean nematode abundance peaked in July, and was lowest in June. This finding is contrary to a report by Vidaković and Bogut (2004) who found nematode abundance in a eutrophic system to be lowest in July.

Seasonal prevalence and abundance also followed similar patterns (Fig. III-3 & III-4, respectively). Both prevalence and mean abundance were lowest in winter and highest in autumn. Spring and summer abundances were significantly higher than in winter, but significantly lower than in autumn. Other studies reported nematode abundance to be highest in summer (Vidaković and Bogut 2004, Traunspurger 1996a, Nalepa & Quigley 1983).

A clear deviation from the trend observed in the current study occurred in April where the spike in mean abundance (Fig. III-2) coincided with a spike in both species richness and H’ (Table III-1); however, no remarkable increase in prevalence was observed (Fig. III-1). During this mo, the system was undergoing a transition from winter into spring as evidenced by the steady increase in substratum temperatures observed between these two seasons (Appendix A-1). The consistency in prevalence coupled with an increase in mean abundance suggests that perhaps isolated pools of nematodes were reproducing in March and April, and increasing their respective population sizes while resources (namely food) were likely most abundant. However, the current study did not examine reproductive rates or individual population growth rates, so this assertion remains purely speculative. A large increase in both species richness and Shannon diversity was observed in April (Table III-1), which suggested that a larger, more species-rich nematode cohort inhabited the system at this time. The fact that nine species representing five genera (Monhystera, Monochromadora, Terschellingia,
Semitobrilus, and Prionchulus) were responsible for contributing approximately 80% of the total nematodes observed in April supported this assertion. This differed from March when only three species from as many genera (Terschellingia, Rhabdolaimus, and Dorylaimus) contributed >80% of the nematodes observed. Both species richness and Shannon diversity peaked in spring (Table III-1), a finding that was also reported by Vidaković and Bogut (2004). This spike in mean abundance, richness, and Shannon diversity was followed by an abrupt decrease in all of these measures in May as the system began to transition into summer (Fig. III-2, Table III-1).

As conditions in the system, e.g., substratum temperature (Appendix A-1), presumably began to stabilize with the onset of summer, it is likely that a loss of habitable sites for a large number of different species resulted, thereby allowing the species best adapted for each habitat to predominate. An increase in the dominance of a few species would then coincide with a loss of species richness as many of the nematode species less adapted to this more stable, i.e., less variable, environment likely began to drop out of the system, i.e., either succumbed to local extinction, or moved to areas not presently sampled, e.g., deeper waters. This trend was observed during June and July in the current system. In particular, during both of these months, prevalence, mean abundance, and species richness all increased (Fig. III-1 & III-2, Table III-1), while Shannon diversity (or evenness) decreased. This is indicative of rearrangement of the nematode community structure such that fewer species were contributing more equally to the total nematode abundance in June, while more species contributed less equally to the total abundance in July, i.e., some species accounted for more of the recovered nematodes than others. Evidence for this trend was observed in the fact that nearly 75% of the total
nematodes recovered in July were contributed by only five species belonging to two genera (Monhystera and Itonchus).

A noticeable decrease in prevalence and mean abundance (Fig. III-1 & III-2, respectively), coupled with an increase in the Shannon diversity index, but constant species richness, was observed in August (the final summer mo) (Table III-1). This is indicative of a decrease in the population size of some of the more abundant nematode species, and an increased degree of isolation, i.e., decreased prevalence, of nematode populations. In other words, all 19 species observed in August were contributing more equally to the total nematode abundance, and were observed in a fewer number of sites than in July. Specifically, five of the most abundant genera (Monhystera, Monochromadora, Anaplectus, Semitobrilus, and Itonchus) observed in July (representing 10 species) showed substantial decreases in abundance (between 41.7 and 100%) in August, and only two of the most abundant genera (Neotobrilus and Dorylaimus) in July (represented by 2 species) showed increases in abundance (33.3 and 35.5%, respectively) in August. Because the observed decreases in abundance outweighed the increases in abundance in this mo, there was an overall decrease in abundance and increase in evenness observed near the end of summer.

Finally, as the system transitioned into autumn (Appendix A-1), prevalence and abundance both increased (Fig. III-3 & III-4, respectively), while richness and Shannon diversity both decreased (Table III-1). This pattern suggests that a smaller number of nematode species contributed disproportionately to the relative abundance of nematodes observed, i.e., relatively few nematode species were responsible for providing a large number of nematodes. In particular, 11 genera (15 species) were responsible for
contributing >10% of the relative abundance in August (the final summer mo), whereas only six genera (10 species) were observed in large enough numbers to account for >10% of the relative abundance in September. The prevalence spike during the latter mo also suggests that as nematode communities increase in number, they disperse and inhabit more sites. Such expansion may have contributed to the observed loss of diversity as the more abundant species began to occupy prime habitat while competitively excluding other, less abundant species. Further, it is possible that this peak (and the one observed in April) coincided with an increase in the number of transects containing algae.

Algae serve as a source of food and potential habitat for a number of nematode species (Peters et al. 2007a), so the presence of these primary producers may influence nematode abundance. However, the preliminary results of the current study do not necessarily implicate a positive influence of algae on the presence or abundance of nematodes as a whole, since both prevalence and mean abundance were higher in mixed substratum than in the mud/algae substratum (Fig. III-5 & III-6, respectively). Algae were rather sporadic in their distribution around the lake over the course of the study and because substratum classifications were made at the beginning of the study, and were not changed at any point during the course of the study, the mud/algae transects may not have been dominated by algae during all seasons. Likewise, non-algae transects may have contained sections of algae at some point during the year.

A final shift in the nematode community composition was observed in the final mo of autumn (September) considered in the current study. In particular, mean abundance and prevalence both declined (Fig. III-2 & III-1, respectively), suggesting that after the population boom observed for multiple nematode species in August, population
sizes began to once again decline. Species richness remained constant, indicating that little change occurred in terms of the number of species present; however, Shannon diversity decreased, suggesting that there was an even greater disparity in the number of nematodes contributed by the species persisting in the system. In other words, the same number of species was still present, but some of them appeared in larger numbers than others, thereby decreasing evenness.

**Influence of substrata**

Prevalence values based on substratum type were very similar (Fig. III-5) and did not differ significantly. This suggests that all substrata were equally capable of supporting the nematofauna in the system, i.e., no one substratum type appeared to be more or less habitable than the others. However, the fact that mean abundance was significantly higher in mixed substratum suggests that it might have been the preferred substratum type (Fig. III-6). Further support of this preferential selection of substratum type was reported by Ullberg and Ólafsson (2003) who found that once nematodes were suspended in the water column, they were capable of actively selecting habitat. One contributing factor to the increased mean abundance in mixed substratum was the fact that two samples from this substratum type contained $\geq 50$ nematodes, which was much higher than the number observed in most samples.

Similarity in species richness estimates for all substrata (ranging from 27-30) further supports the assertion of near equal habitability of these substrata by nematodes in general. Furthermore, the ability of nematodes to disperse via water-column transport contributes to nematode distribution on larger scales (Peters et al. 2005). This likely accounts for the similarities in community structure maintained in the various substrata,
even though many patches of habitat containing the same substratum were separated by large distances.

It is important to note that not all substrata may have been sampled with sufficient effort as evidenced by the failure of the substratum type species accumulation curves to reach plateaus based on the number of samples observed (Appendix D-3). The apparent lack of the accumulation curves to reach asymptotes suggests that the observed richness may not reflect the true richness in this system. This is most clearly represented in the curve for leaf substratum, as it appeared to be the most distant from a plateau. Additionally, the curve actually crossed over all other curves, suggesting that the richness estimate in this substratum type is more influenced by the number of samples observed than were the other substrata. This is not entirely surprising due to the fact that a greater proportion of transects in the current system contained leaf substratum than any other substratum sampled. As a result, the likelihood of re-sampling leaf transects was greatly reduced when compared to the other substrata types, thereby making it increasingly more difficult to estimate the true species richness in this substratum type.

Interestingly, mean abundance was lower in leaf substratum than in any other substrata (Fig. III-6), but this difference was not significant when compared to mud/clay and mud/algae substrata. This discrepancy indicates that either the methodology utilized in the current study was not adequate for collecting nematodes from this substratum type, or that perhaps this substratum type was a transitory habitat for the nematodes in the system. In other words, it was capable of supporting a wide variety of nematodes (greater richness), but fewer individuals (lower abundance). Based on the similarity of prevalence and Shannon diversity values attributed to leaf substratum relative to the
values observed in the other substrata, it is likely that the latter explanation is most appropriate. In general, it appeared that, at least on the scale of the entire system, substratum type (when compared to the influence of seasonality) had relatively little impact on structuring the nematode community.

A sampling approach that focused on re-sampling the same transects on multiple occasions may help elucidate the influence of these factors on a smaller microhabitat scale. Such information would be beneficial because it would allow for finer resolution of what is occurring on the local community (and perhaps even the population) scale, rather than the larger, global scale observed in this study. The ability to view how local communities change based on the influence of season, and particularly substratum type, would better help to explain the patterns observed in this study. With the exception of leaf substratum on the N and W banks of the lake, there was typically anywhere from tens to hundreds of m separating transects containing the same substratum. A closer look at the dispersal ability of various nematodes in this system could provide answers to questions concerning how transects containing the same substratum type, but that are separated by rather large distances, still appear to contain similar nematode community compositions. It seems most probable that the patterns in community structure observed based on substratum type are driven by a combination of the hypothetically long-distance passive dispersal ability of free-living nematodes in the water column (Hagerman and Rieger 1981, Palmer 1988, Atilla and Fleeger 2000), coupled with their ability to selectively inhabit particular substrata, i.e., habitats (Ullberg and Ólafsson 2003). Furthermore, a population-genetics approach could support both long distance, passive dispersion, and the similarity in arrangement of the community.
LITERATURE CITED


Eskin RA & Palmer MA  1985.  Suspension of marine nematodes in a turbulent tidal

Gomes GS, Huang SP, & Cares JE  2003.  Nematode community, trophic structure, and
population fluctuation in soybean fields.  Fitopatol. Bras.  28: 258-266.

Hagerman GM & Rieger RM  1981.  Dispersal of benthic meiofauna by wave and current

Hodda M & Eyuale-Abebe  2006.  Techniques for processing freshwater nematodes. In:
Freshwater nematodes:  ecology and taxonomy.  Eyuale-Abebe, Traunspurger

Ingham RE, Tofymow JA, Ingham E, & Coleman DC  1985.  Interactions of bacteria,
fungi, and their nematode grazers: effects on nutrient cycling and plant growth.

University Press, Princeton.

Michiels IC & Traunspurger W  2004.  A three year study of seasonal dynamics of a

Nalepa TF & Quigley MA  1983.  Abundance and biomass of the meiobenthos in
nearshore Lake Michigan with comparisons to the macrobenthos.  J. Great Lakes
Res.  9: 530-547.

explaining passive transport and active emergence with implications for


Table III-1: Nematode prevalence, mean abundance, and diversity measures observed for each mo, season, and substratum type. Mean abundance = mean (± SE), S = species richness, $H'$ = Shannon-Wiener diversity index. Nematodes were not morphotyped in January or February so no diversity measures could be calculated.

<table>
<thead>
<tr>
<th></th>
<th>Prevalence (%)</th>
<th>Mean Abundance (± SE)</th>
<th>S</th>
<th>$H'$</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Overall</strong></td>
<td>65.7</td>
<td>4.9 (± 0.5)</td>
<td>45</td>
<td>2.5</td>
</tr>
<tr>
<td><strong>Month</strong></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>January</td>
<td>75.0</td>
<td>3.0 (± 0.8)</td>
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<tr>
<td>February</td>
<td>39.6</td>
<td>1.7 (± 0.6)</td>
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<td>--</td>
</tr>
<tr>
<td>March</td>
<td>54.2</td>
<td>1.7 (± 0.7)</td>
<td>10</td>
<td>1.5</td>
</tr>
<tr>
<td>April</td>
<td>55.9</td>
<td>5.7 (± 1.3)</td>
<td>29</td>
<td>2.4</td>
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<tr>
<td>May</td>
<td>45.8</td>
<td>1.9 (± 0.9)</td>
<td>17</td>
<td>1.6</td>
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<tr>
<td>June</td>
<td>47.8</td>
<td>1.0 (± 0.3)</td>
<td>14</td>
<td>1.9</td>
</tr>
<tr>
<td>July</td>
<td>91.7</td>
<td>10.9 (± 2.9)</td>
<td>19</td>
<td>1.8</td>
</tr>
<tr>
<td>August</td>
<td>81.3</td>
<td>5.2 (± 0.8)</td>
<td>19</td>
<td>2.0</td>
</tr>
<tr>
<td>September</td>
<td>89.6</td>
<td>8.9 (± 1.7)</td>
<td>15</td>
<td>1.9</td>
</tr>
<tr>
<td>October</td>
<td>79.2</td>
<td>5.8 (± 1.4)</td>
<td>15</td>
<td>1.8</td>
</tr>
<tr>
<td><strong>Season</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Winter</td>
<td>48.8</td>
<td>2.0 (± 0.5)</td>
<td>10</td>
<td>1.5</td>
</tr>
<tr>
<td>Spring</td>
<td>54.2</td>
<td>4.1 (± 0.8)</td>
<td>32</td>
<td>2.4</td>
</tr>
<tr>
<td>Summer</td>
<td>69.3</td>
<td>5.1 (± 0.9)</td>
<td>29</td>
<td>2.2</td>
</tr>
<tr>
<td>Autumn</td>
<td>84.4</td>
<td>7.3 (± 1.1)</td>
<td>19</td>
<td>2.0</td>
</tr>
<tr>
<td><strong>Substratum Type</strong></td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Leaf</td>
<td>66.7</td>
<td>3.9 (± 0.7)</td>
<td>30</td>
<td>2.2</td>
</tr>
<tr>
<td>Mixed</td>
<td>76.3</td>
<td>9.4 (± 2.0)</td>
<td>29</td>
<td>2.0</td>
</tr>
<tr>
<td>Mud-Algae</td>
<td>61.4</td>
<td>4.3 (± 0.7)</td>
<td>27</td>
<td>2.3</td>
</tr>
<tr>
<td>Mud/Clay</td>
<td>63.0</td>
<td>5.7 (± 0.9)</td>
<td>28</td>
<td>2.2</td>
</tr>
</tbody>
</table>
Fig. III-1: Monthly prevalence of free-living nematodes.
The diagram shows the prevalence (%) over different months. The trend line indicates a positive correlation, with an $R^2$ value of 0.3584.
Fig. III-2: Monthly mean abundance of free-living nematodes. Error bars are SE. Monte Carlo simulation analyses were used to determine whether mo differed significantly from one another. P-values can be found in Appendix B-1.
Fig. III-3: Seasonal free-living nematode prevalence.
**Fig. III-4:** Seasonal free-living nematode mean abundance. Error bars are SE. Asterisks denote significant differences from all other seasons. Monte Carlo simulation analyses were used to determine whether seasons differed significantly from one another. P-values can be found in Appendix B-2.
Mean Nematode Abundance

Season

Winter Spring Summer Autumn

R² = 0.9202
Fig. III-5: Prevalence of free-living nematodes based on substratum type.
**Fig. III-6:** Mean abundance of free-living nematodes based on substratum type. Error bars are SE. Asterisk denotes a significant difference. Monte Carlo simulation analyses were used to determine whether substrata differed significantly from one another. P-values can be found in Appendix B-3.
CHAPTER IV

The Role of Spatiotemporal Influences on the Prevalence, Mean Abundance, and Overall Community Dynamics of Free-living Nematodes Classified by Feeding Type in a Freshwater System

ABSTRACT

Few studies have attempted to better understand the influence of abiotic and biotic factors on the composition of the free-living nematode communities in freshwater systems. The present study was undertaken to discern the role of spatiotemporal factors, i.e., season and substratum type, on the community composition of the free-living nematofauna in a freshwater system. Substratum samples were collected from January through October, 2009 from Mallard Lake (a 4.9 ha, eutrophic pond located in the Piedmont region of North Carolina). Nematodes were recovered from substratum samples via the Baermann extraction method, and were isolated, identified, and quantified. In an attempt to achieve finer resolution of the observed fluctuations in community composition in the system, all identified nematodes were classified into groups based on feeding type. Five such groups were observed: (1.) herbivores, (2.) bacterivores, (3.) unicellular eukaryote feeders, (4.) predators of animals, and (5.) omnivores. Prevalence and mean abundance values were calculated based on feeding type for (1.) the entire system, (2.) each season, and (3.) each substratum type observed.

A total of 1,918 nematodes was recovered, representing 44 species and 29 genera. Of the total nematodes collected, 51.8% were classified as bacterivores, 21.3% were
predators, 14.2% were omnivores, 11.3% were unicellular eukaryote feeders, and 1.3% were herbivores. *Monhystera* were the dominant bacterivores, *Iotonchus* were the dominant predators, *Dorylaimus* were the dominant omnivores, *Semitobrilus* and *Neotobrilus* were the dominant unicellular eukaryote feeders, and *Lelenchus* sp. were the dominant herbivores. Both season and substratum type appeared to influence the community composition in this system, with season showing the greatest influence on the predator feeding type, and substratum type showing the greatest influence on predators and omnivores. Bacterivores were dominant in nearly every season and every substratum type, and showed little fluctuation in either prevalence or mean abundance.
INTRODUCTION

Relatively few studies exist that attempt to explain the influence of spatiotemporal factors on the community composition of the free-living nematofauna in freshwater systems. Still fewer studies attempt to do so in a eutrophic system. Those that do exist appear to be mixed in their conclusions. On the one hand, several studies suggest that both spatial (Prejs 1977, Vidaković and Bogut 2004, Bert et al. 2007) and temporal factors (Vidaković and Bogut 2004) influence free-living nematodes assemblages. Conversely, Michiels and Traunspurger (2004) reported that seasonality had very little influence on the free-living nematode community in a eutrophic lake, Lake Obersee, in Germany. The current study provides a rare look at the role of both of these factors in a eutrophic system. Such studies, exploring the influence of both spatial and temporal variables on the community structure of free-living nematodes, are capable of providing answers to questions that could not be determined by examining either factor alone (Gomes et al. 2003).

In the current study, nematode prevalence and mean abundance were used as measures of the influence of season and substratum type on these meiobenthic organisms. Nematodes were collected from the substratum, recovered via Baermann extraction techniques, isolated, identified to genus, and quantified. Nematodes were then placed into feeding groups based on the type of food, e.g., bacteria, algae, animals, or plants, primarily consumed by each genus. Feeding type assignments followed those proposed by Yeates et al. (1993). It was assumed that the freshwater nematodes observed in this system utilized the same primary nutrient sources as their soil dwelling counterparts.
described in Yeates et al. (1993). This assumption is in agreement with Moens et al. (2006). Genera observed in this system, but not described in Yeates et al. (1993), were assigned to the feeding type most commonly observed for other genera in the same family, when possible.

Five of the eight feeding types described by Yeates et al. (1993) were observed in the current system. These feeding types were herbivore, bacterivore, unicellular eukaryote feeder, predator of animals, and omnivore. Individuals representing the remaining three classifications, i.e., hyphal feeders, substrate ingesters, and dispersal or infective stages of animal parasites, were not observed. One parasitic order (Mermithida) was seen, but these individuals did not fit into the current feeding classification and, therefore, were not included in these analyses.

Yeates et al. (1993) described the food items of these various feeding types in some detail. Herbivores, or plant feeders, feed on vascular plants and use either a tylenchoid stomatostyle or dorylaimoid odontostyle to do so. This feeding type includes both polyphagous or host specific nematodes, as well as epidermal cell or root hair feeders, and algal feeders that feed through a narrow stylet (and, therefore, do not swallow unicellular algae intact). Bacterivores, or bacterial feeding nematodes, include any species that ingest prokaryotes as their primary food source, and do so either through a stylet or with a broad mouth. Unicellular eukaryote feeders ingest diatoms, algae, fungal spores, and whole yeast cells. The animal predators feed on a range of invertebrates, e.g., protozoa, nematodes, rotifers, and enchystraeids, and can be classified as either ingesters or piercers, depending on the manner in which they consume their prey (Table IV-1). Piercers bear a narrow stylet through which they suck the body fluids of
their prey items. Mononchids, which were the most common predator in the current system, are extreme prey generalists (Bilgrami et al. 1986, Khan and Kim 2007), where detection of prey requires direct contact (Nelmes 1974, Bilgrami et al. 1984). Omnivores feed on a wide range of food items, e.g., fungal hyphae, bacteria, various invertebrates, diatoms, algae, fungal spores, and yeast cells, and individuals possessing a stylet may also feed on plant tissues. Several studies suggest that of these food items, algae, fungi, and nematodes are perhaps the most commonly consumed (Hollis 1957, Ferris 1968, Wood 1973). Furthermore, at least some dorylaimid species, e.g., *Dorylaimus stagnalis*, are predacious upon chance encounters (Linford and Oliveira 1937), and do not appear to be attracted to live or excised prey items (Shafqat et al. 1987). However, Shatgat et al. (1987) reported that dorylaimids also aggregate at feeding sites, suggesting that perhaps some level of chemosensory response is possible.

The purpose of the current study was multifaceted. First, a resolution of the contribution of various feeding types to the overall nematode community in the freshwater system was desired. Second, the current study sought to determine what influence spatiotemporal factors had on the patterns of prevalence and abundance for the feeding types present. Finally, the study was interested in understanding the trophic relationships that existed between the observed nematode feeding types.
MATERIALS AND METHODS

Sampling method

Samples were collected from Mallard Lake according to the methods described in Chapter III.

Mo were grouped into seasons based on naturally occurring breaks in substratum temperature (Appendix A). Winter consisted of collections obtained on March 26, spring collections took place from April 2 through May 8, summer collections from May 28 through August 18, and autumn collections from September 10 through October 28, with all sampling taking place in 2009.

Nematode recovery

Nematodes were recovered using a Baermann-funnel apparatus, identified, and stored as described in Chapter III.

Feeding type classification

Nematode morphotypes were identified to genus based on morphological characteristics. Classification was based on the list proposed by De Ley and Blaxter (2004). To ensure consistency when morphotyping recovered nematodes, detailed drawings, photographs, and morphological descriptions were created for each morphotype observed. Once morphotypes were assigned to a genus, each taxon was assigned to a feeding type according to Yeates et al. (1993). This classification system was based on the primary food source utilized by each genus (or family when genus-level information was not provided). According to Yeates et al. (1993), soil nematodes can be placed into eight different feeding type classifications: (1.) plant feeders, (2.) hyphal
feeders, (3.) bacterial feeders, (4.) substrate ingesters, (5.) predators of animals, (6.) unicellular eukaryote feeders, (7.) dispersal or infective stages of parasites, and (8.) omnivores. For the purposes of this study, it was assumed that individuals belonging to the same genus (or family) obtained nutrients from the same source in both soil and aquatic habitats.

Data analysis

Of the 2,349 total nematodes collected, only 1,828 were identified to morphotype. This discrepancy was the result of 1 of 3 factors: (1) morphotyping did not begin until the 26 March 2009 collection, so the identity of any nematodes collected prior to this date is unknown, (2) some nematodes were too small to be clearly identified, or (3) a very small number of nematodes were lost during transfer from the stereomicroscope to the bright field microscope. Furthermore, of the 1,828 nematodes that were successfully morphotyped and taxonomically classified, only 1,818 nematodes were classified into feeding types based on those proposed by Yeates et al. (1993). The 10 nematodes not included in this feeding type classification belonged to genera (and families) not described by Yeates et al. (1993).

Because individual identification of nematodes did not begin until the final sampling event in March 2009, the data set for this mo (and consequently winter as a whole) consisted of only 12 independent samples. This oversight is mentioned here in the spirit of full disclosure, and was considered when analyzing these data. Feeding-type prevalence and mean abundance values were calculated both seasonally and based on substratum type. Multiple regression analysis was utilized to determine whether a significant relationship existed between prevalence and season across all feeding types,
and individual regression analyses were performed to determine whether the relationship between prevalence and season for each individual feeding type was significant.

Two series of Chi-square analyses were conducted on the feeding type data based on season and substratum type. The first examined each season or substratum type separately, and sought to determine whether significant differences existed between observed and expected prevalence values for all 5 feeding types, i.e., data classes. The observed numbers for each data class were simply the number of times each feeding type was observed, i.e., presence, in each season or substratum type. Expected values for these analyses were generated by multiplying the sum of the observed presence values for all feeding types, times the proportion of each feeding type actually observed in the system as a whole, i.e., the prevalence of each feeding type (see Chapter III). Because the percent contribution of each feeding type to the overall abundance observed in the system was known, these proportions could be used to generate the expected values for each data class, i.e., the number of times one would expect to observe each feeding type in each season or substratum type based on what was actually observed in the system.

The second series of Chi-square analyses sought to determine whether observed prevalence values differed significantly from expected values between seasons or substrata for each feeding type. Therefore, each feeding type was analyzed separately, and the data classes considered in these analyses were either seasons or substrata. Observed values for each data class corresponded to the observed presence data for individual feeding types in each season or substratum type. The expected proportions utilized in these analyses were based on the proportions of all the nematodes in the system found in each season or substratum type, i.e., the calculated nematode prevalence
obtained for each season or substratum type in the system as a whole (see Chapter III). The expected number for each data class, i.e., season or substratum type, was generated by multiplying the summed seasonal or substrata presence values for each feeding type, and the proportion of nematodes found in each season or substratum type for the whole system (see Chapter III). Further discussion of trends for both series of Chi-square analyses, i.e., determining which feeding types, seasons, or substrata deviated the most from expected values, was purely observational, and was not analyzed statistically due to the subsequent loss of statistical power associated with conducting multiple Chi-square analyses with low degrees of freedom.

Differences in mean abundance were considered significant when no overlap of standard error was observed between, or among, seasons or substrata. The significance threshold for all analyses was set at $p \leq 0.05$.

**Terminology**

The term relative abundance is defined in the context of the current study as the percent of the total nematofauna contributed by any one feeding type or genus. The use of the terms prevalence, mean abundance, sampling units, morphotypes, and genus names were described in Chapter III.
RESULTS

Overall nematode assemblage based on feeding type

A total of 1,918 nematodes was identified to genus level based on morphology. Five of the eight feeding types described in Yeates et al. (1993) were observed in the current system: plant feeders, bacterial feeders, predators of animals, unicellular eukaryote feeders, and omnivores. In the current study, these feeding types were referred to as herbivores, bacterivores, predators, unicellular eukaryote feeders, and omnivores, respectively. Bacterivores were the most prevalent (Table IV-2), the most abundant (Fig. IV-3), and the most speciose feeding type observed in this system. This feeding type composed over half of the total nematode community (51.8%), and was represented by 26 species in 19 genera (Table IV-3). One genus, Monhystera, contributed 68.8% of these individuals.

Predators were the second most abundant feeding type (Fig. IV-3), contributing 21.3% of the total nematodes observed (Table IV-3), and the second most speciose feeding type (8 species in 6 genera). This was, however, the fourth most prevalent feeding type, and was recovered from 17.1% of the total samples (Table IV-2). The predator feeding type was dominated by prey-ingesting predators (99.2%), which included species of Iotonchus and Prionchulus (Table IV-3), among others (though in greatly reduced numbers). Only 0.08% of predators were prey-piercers and all of these individuals belonged to Seinura.

A single genus, Dorylaimus, represented the omnivore feeding type and made up 14.2% of the total nematode community (Table IV-3). Omnivorous individuals were observed in 29.4% of samples, making them the second most prevalent feeding type in
this system (Table IV-2). Unicellular eukaryote feeders made up 11.3% of the total nematode fauna, and were represented in nearly equal proportions by individuals belonging to two genera, *Semitobrilus* (51.2%) and *Neotobrilus* (48.8%) (Table IV-3). This was the third most prevalent feeding type, occurring in 21.3% of samples (Table IV-2). Herbivores were the least abundant (Fig. IV-3) and the least prevalent (Table IV-2) feeding type observed. A single genus, *Lelenchus*, was the only herbivorous nematode present in this system.

**Prevalence of feeding types**

*Season:* A positive trend in prevalence was observed for all feeding types such that in autumn, prevalence values were higher than in winter (Table IV-2, Fig. IV-1). No significant relationship was observed between prevalence and season when all feeding types were considered together ($R^2=0.691$, $p=0.169$). However, when each feeding type was considered separately, a significant positive relationship was observed for bacterivores ($R^2=0.999$, $p<0.001$), unicellular eukaryote feeders ($R^2=0.961$, $p=0.020$), and omnivores ($R^2=0.959$, $p=0.021$). Although these three feeding types all exhibited a seasonal increase in prevalence, the number of sites occupied by unicellular eukaryote feeders and omnivores rarely exceeded 50% of the number of sites occupied by bacterivores (Table IV-2). The relationship between prevalence and season was not significant for herbivores or predators ($R^2=0.648$, $p=0.195$ and $R^2=0.852$, $p=0.077$, respectively).

Observed presence did not differ significantly from expected in either winter or spring ($\chi^2$ test, $p=0.483$ and $\chi^2$ test, $p=0.097$, respectively). However, these values were significantly different than expected values in summer and autumn ($\chi^2$ test, $p<0.001$ and
During summer, unicellular eukaryote feeders and omnivores were observed in more samples than expected, while predators were observed in fewer samples than expected. In autumn, bacterivores were recovered from fewer samples than expected, while herbivores and omnivores were observed in more samples than expected. Thus, omnivores were more prevalent in both summer and autumn than expected.

Bacterivores were more prevalent than any other feeding type in all seasons (Table IV-2), and observed presence differed significantly from expected among the seasons ($\chi^2$ test, $p<0.001$). The largest discrepancy for this feeding type was observed in winter, when it was recovered in fewer samples than expected. Based on season, all other feeding type presence values differed significantly from the expected prevalence values as well ($\chi^2$ test, $p \leq 0.032$ for all feeding types), and a description of the patterns follows.

A very clear transition in the community composition of unicellular eukaryote feeders and omnivores was observed as the system changed from spring into summer (Table IV-2, Fig. IV-1). In particular, unicellular eukaryote feeders were more prevalent than omnivores during the winter and spring mo, and less prevalent than omnivores during the summer and autumn mo. Although the prevalence of both of these feeding types increased steadily throughout the study, omnivore prevalence increased much more rapidly from spring to autumn than did that of unicellular eukaryote feeders. Observed presence values differed significantly from expected values for unicellular eukaryote feeders ($\chi^2$ test, $p=0.032$), with the largest discrepancy occurring in winter when this feeding type was recovered from fewer sites than expected. Similarly, the presence of omnivores differed significantly from what was predicted based on observed patterns in
the overall system ($\chi^2$ test, $p<0.001$), with the greatest discrepancies occurring in winter and autumn. Specifically, this feeding type was recovered from fewer sites than expected in winter, and from more sites than expected in autumn.

Observed predator presence differed significantly from expected among seasons ($\chi^2$ test, $p<0.001$). Similar to the trend observed for omnivore, predators were recovered from fewer sites in winter and more sites in autumn than predicted. Furthermore, predator prevalence ranged from 0% in the winter to 33.3% in autumn (Table IV-2, Fig. IV-1). Herbivores were consistently observed in fewer sites (lower prevalence) than any other feeding type throughout the entire course of the study (Table IV-2, Fig. IV-1).

During the winter season both herbivores and predators were absent from the system. Herbivore presence in the system differed significantly from what was predicted ($\chi^2$ test, $p=0.001$) and, similar to the pattern observed for predators, herbivores showed a notable spike in prevalence between summer and autumn. This increase amounted to a 3-fold jump in prevalence for predators, and a nearly 10-fold jump in prevalence for herbivores. Thus when compared to expected values, unicellular eukaryote feeders, omnivores, predators, and herbivores were all notably absent from the system in winter, and omnivores, predators, and herbivores had higher presence values in autumn.

Substratum type: Bacterivores were the most prevalent feeding type observed in all substrata (Table IV-2, Fig. IV-2). Prevalence of bacterivores, omnivores, and predators all followed similar trends. When the prevalence of one increased or decreased, so too did the prevalence values of the other two feeding types. Prevalence was highest for all three in mixed and mud/clay substrata, and lowest in leaf and mud/algae substrata. In all cases, bacterivores and omnivores were more prevalent than predators. The observed
presence of bacterivores did not differ significantly from expected values ($\chi^2$ test, $p=0.315$); however, the observed presence of omnivores and predators did ($\chi^2$ test, $p<0.001$ and $\chi^2$ test, $p<0.001$, respectively). Omnivores were observed in more samples than expected in the mixed substratum, both omnivores and predators were recovered from fewer samples than expected in the mud/algae substratum, and predators were observed in a greater number of sites than predicted in mud/clay substratum.

Herbivores were the least prevalent in all substrata (Table IV-2, Fig. IV-2). There was a relatively large amount of variability in prevalence values observed in this feeding group, but observed presence values did differ significantly from those predicted based on the system as a whole ($\chi^2$ test, $p=0.001$). Herbivore prevalence peaked at 9.7% in mud/clay substratum, where it was nearly equivalent to the prevalence of unicellular eukaryote feeders (Fig. IV-2). Interestingly, this peak corresponded to the maximum observed predator prevalence (33.3%), and one of the equally high maxima observed for omnivores (46.2%). The other maximum in omnivore prevalence occurred in mixed substratum. Substratum type preference was most distinct in the omnivore feeding type (Fig. IV-2). Prevalence was highest for bacterivores (59%) and unicellular eukaryote feeders (29.5%) in the mixed substratum. Regardless of this peak in prevalence, the observed presence of unicellular eukaryote feeders did not differ significantly from expected values among substrata ($\chi^2$ test, $p=0.088$).

Feeding type presence differed significantly from expected presence values based on each feeding type’s contribution to the nematode community as a whole ($\chi^2$ test, $p \leq 0.007$ in all substrata). A closer look at these data revealed that in the leaf substratum, unicellular eukaryote feeders were found in nearly twice as many samples as expected,
while predators were found in fewer samples than predicted. In the mixed substratum, unicellular eukaryote feeders and omnivores were observed in more samples than expected, while bacterivores and predators were observed in fewer samples than expected. In the mud/algae substratum, unicellular eukaryote feeders were present in more samples than predicted, while predators were present in fewer than predicted. Finally, in autumn, bacterivores were observed in a fewer number of sites than predicted, while herbivores were observed in nearly 9 times more samples than expected, and omnivores were recovered from nearly twice as many sites as predicted.

Abundance of feeding types

Season: Bacterivores were the dominant feeding type during all seasons (Fig. IV-4), with winter being the only season in which this dominance was not significant. During this period, bacterivores and omnivores were observed in nearly equivalent abundances. A clear bimodal pattern in seasonality was observed based on the mean abundance values for the primary consumers (bacterivores, unicellular eukaryote feeders, and herbivores) in the present study. Peaks were observed in spring and autumn, and troughs were observed in winter and summer. The secondary consumers in the system (predators and omnivores) showed a different pattern in mean abundance, with omnivores increasing from spring to autumn, and predators increasing from winter to autumn. With the exception of unicellular eukaryote feeders (who reached a maximum in spring), maximum mean abundance for all feeding types was observed in autumn. In all cases, the mean abundance values observed in autumn were significantly higher than those observed in spring (Appendix A-3). Even though most feeding types showed steady increases in mean abundance over the course of the study, there was not a significant relationship
between mean abundance and season when all feeding types were considered together ($R^2=0.420$, $p=0.352$).

Comparison of standard errors showed that bacterivores and herbivores were significantly more abundant in autumn than in winter and summer, but differences in mean abundance between autumn and spring were not significant (Appendix A-3). A similar trend was observed for unicellular eukaryote feeders in which their mean abundance was highest in spring. Although it was not significantly different from autumn, mean abundance in spring was significantly different than the values obtained in winter and summer (Appendix A-3). Omnivores followed a different pattern, showing peaks in winter and autumn, and a trough in spring and summer (Fig. IV-4). The variability in mean abundance for this feeding type was not statistically significant in winter, spring, and summer; although the peak in abundance observed in autumn was significantly different than their relatively low mean abundances in spring and summer (Appendix A-3). Differences between winter and autumn mean abundance were not significant for omnivores.

Predators (like herbivores) were completely absent from the system during the winter mo; however, unlike herbivores, mean predator abundance showed a steady increase from spring to autumn (Fig. IV-4). Predators were significantly more abundant in autumn than in winter or spring, but the difference between summer and autumn was not significant. Further, there was a significant relationship between mean abundance and season for predacious nematodes ($R^2=0.993$, $p=0.004$). This was the only feeding type that showed this significant relationship based on independent regression analyses of all feeding types ($R^2 \leq 0.707$, $p \geq 0.159$ for all other feeding types).
Species of *Monhystera* exhibited the highest relative abundance of all bacterivores in spring, summer, and autumn (43.4, 74.6, and 91.1%, respectively). *Terschellingia* had the highest relative abundance during the winter. The bacterivore *Monochromadora* was observed in winter, spring, and summer, but with consistently lower relative abundance values than *Monhystera*. *Semitobrilus* and *Neotobrilus* dominated the unicellular eukaryote feeding type. Indeed, *Semitobrilus* had the highest relative abundances for this feeding type in winter and spring (100 and 88.5%, respectively), whereas *Neotobrilus* had the highest relative abundances for this feeding type in summer and autumn (49.2 and 98.4%, respectively).

The only strictly herbivorous genus observed was *Lelenchus* and it occurred in consistently low abundance relative to the other genera. The only genus classified as omnivorous was *Dorylaimus*, and it was found in high abundance relative to other genera in both summer and autumn (17.4 and 16.7%, respectively). It had the third highest relative abundance in both of these seasons. The dominant predators during all seasons included *Prionchulus*, *Iotonchus*, and *Ironus*. *Prionchulus* had the highest relative abundance among nematodes of this feeding type during spring (82.5%), but this dominance switched to *Iotonchus* in both summer (90.9%) and autumn (83.1%). Finally, *Iotonchus* exhibited the second highest relative abundance across all genera, second only to *Monhystera*, in both summer (23.3%) and autumn (19.8%).

*Substratum type:* Bacterivores were significantly more abundant than all other feeding types in all substrata except mixed, where they occurred in the same numbers as predators (Fig. IV-5). Additionally, no significant differences were observed in mean abundance of bacterivores across substrata (Fig. IV-6). Herbivores were significantly
less abundant than all other feeding types in all substrata, except mud/clay where they occurred in nearly equal abundance as predators (Fig. IV-5). Herbivores were the only feeding type significantly more abundant in the mud/clay substratum than in any other substrata.

Unicellular eukaryote feeders occurred with the greatest abundance in mixed substratum, although this peak was not significantly different than the mean abundance values observed in leaf and mud/clay substrata (Fig. IV-5). The difference in means was significant between mixed and mud/algae substrata (Fig. IV-6). Omnivores were significantly less abundant in leaf substratum than in any other substratum, and significantly more abundant in mixed substratum than mud/clay substratum. Neither of these substratum types differed significantly from mud/algae substratum (Fig. IV-6). Predators were significantly more abundant in mixed than any other substrata, occurred in nearly the same numbers in leaf and mud/algae, and were significantly less abundant in mud/clay substratum than any other substrata. This peak abundance of predators in mixed substratum appeared to correspond to peaks in omnivore and unicellular eukaryote feeder abundance in the same substratum type, although differences in mean abundance between substrata within each of these two feeding types were much less obvious than those observed for predators (Fig. IV-5). For instance, mean abundance peaked in mixed substratum for bacterivorous and unicellular eukaryote feeding types; however, it was not significantly different than values observed in any of the other substrata (Fig. IV-6). The single exception pertained to omnivores in the leaf substratum as reported above.

An additional trend in nematode community composition was observed in mixed, mud/algae, and mud/clay substrata. In these three substrata, bacterivores were always the
most dominant, omnivores were always the second most dominant, unicellular eukaryote feeders were third, and herbivores were the least abundant in all three substrata (Fig. IV-5). Predators did not follow a consistent trend in these substrata.

*Monhystera* exhibited the highest relative abundance of any of the bacterivores in all substrata, ranging from 63.2% in mud/algae substratum to 80.6% in leaf substratum. Other notable genera contributing to the overall abundance of bacterivores included *Monochromadora, Thalassomonhystera, Terschellingia, Eumonhystera,* and *Rhabdolaimus,* although in all cases, these genera contributed fewer individuals than *Monhystera.* In all substrata, the unicellular eukaryote feeders with the highest relative abundance belonged to *Semitobrilus* and *Neotobrilus.* *Semitobrilus* had the highest relative abundance in mud/algae (70.0%) and mud/clay (77.4%) substrata, whereas *Neotobrilus* had the highest relative abundance in leaf (62.3%) and mixed (74.2%) substrata. The herbivore *Lelenchus* was only observed in mixed, mud/algae, and mud/clay substrata, and typically occurred with lower abundance than did any of the other genera in any substrata. The omnivore *Dorylaimus* was found in all substrata, but had the highest relative abundance values in mixed, mud/algae, and mud/clay substrata, where they contributed 15.1, 16.9, and 19.2%, respectively, of nematodes.

Two predator genera, *Iotonchus* and *Prionchulus,* had the highest relative abundances in this feeding type. Specifically, *Iotonchus* had the highest relative abundance of all predators in leaf (53.4%), mixed (93.7%), and mud/algae substratum (57.5%), while *Prionchulus* had the highest relative abundance for all predators in the mud/clay substratum (66.7%). In the mixed substratum, *Iotonchus* had the highest
relative abundance of all genera (35.3%), followed by *Monhystera* (28.1%) and *Dorylaimus* (15.1%).
DISCUSSION

Analysis of the system based on feeding type

In this freshwater system, bacterivorous nematodes comprised 51.8% of the total
nematode community. In addition, this feeding type was significantly more abundant
than all other feeding types present (Fig. IV-1). Predators and omnivores were the next
most abundant, respectively, followed by epistrate feeders and finally herbivores (Fig.
IV-3). The primary consumers (bacterivores, unicellular eukaryote feeders, and
herbivores) were dominated by Monhystera, Semitobrilus, and Lelenchus, respectively.
Monhystera is typically associated with oligotrophic systems (Prejs 1977, Wu & Liang
1999), but was found in high abundance in the current system, which was considered to
be eutrophic. Prejs (1977) and Michiels and Traunspurger (2004) both reported that
nematodes from the family Monhysteridae also appear in large abundances in some
eutrophic systems. Other dominant bacterivorous nematode genera included
Monochromadora, Terschellingia, Thalassomonhystera, Eumonhystera, Prismatolaimus,
and Rhabdolaimus, among others (Table IV-3). The two most common unicellular
eukaryote feeders, Neotobrilus and Semitobrilus, occurred in nearly equal proportions in
this system. Lelenchus was the only herbivorous nematode observed. Secondary
consumers (predators and omnivores) were dominated by Iotonchus and Dorylaimus,
respectively. Another predominant predator, Prionchulus, occurred quite frequently;
however, all other strictly predacious genera were found in very low numbers.
Dorylaimus was the only omnivorous genus observed.
Influence of season and trophic interactions

Season appeared to play an important role in structuring the nematode community in Mallard Lake. A significant relationship between prevalence and season was observed for bacterivores, unicellular eukaryote feeders, and omnivores (Fig IV-1). This suggests that, as the study progressed, nematodes utilizing these feeding types dispersed in the lake, thus inhabiting progressively more sites. Peters et al. (2005) showed that, for hard substrata, habitats that were more attractive to nematodes tended to be either colonized by other nematodes and meiobenthos, or contained a greater amount of periphyton components, e.g., algae, bacteria, fungi, etc., all of which increase on a temporal scale. Although the current study consisted of samples taken from predominantly soft substratum, it seems logical that habitats containing similar components would still be preferred. This selectivity would be most important to the three feeding types that showed significant increases in prevalence in the present study. As time passed, it is logical that periphyton elements also increased, providing more preferential habitat for these feeding types.

In terms of mean abundance, predators appeared to be more influenced by season than any of the other feeding types. This assertion is initially based on the fact that the relationship between mean abundance and season was only significant for nematodes classified as this feeding type. Moreover, predators were significantly more abundant in autumn than they were in winter (Fig. IV-4). In fact, no predatory nematodes were recovered in any of the winter collections, yet, by autumn, it was the second most common feeding type observed. With the exception of omnivores, all feeding types were
significantly more abundant in autumn than in winter, further implicating season as a driving force behind the community structure of free-living nematodes in the system.

When considering the prevalence and mean abundance data for the nematode community as a whole, i.e., all feeding types included, season does indeed play a role in structuring the community. This is clear because both of these measures increased as the study progressed. However, it becomes more difficult to elucidate the role of season when examining the feeding types separately. Because seasonality does not appear to strongly influence all observed feeding types, it seems probable that some other factor closely related to seasonality, e.g., trophic interactions, may be influencing the observed patterns of prevalence and abundance at the level of feeding type.

Two major nematode trophic groups—based on feeding type—were observed in the system: (1.) primary consumers (bacterivores, unicellular eukaryote feeders, and herbivores), and (2.) secondary consumers (predators and omnivores). These two trophic groups appeared to have strong influences on one another over the course of the study. All primary consumers in the study followed the same pattern in mean abundance such that peaks were observed in spring and autumn, and troughs were observed in winter and summer. In addition, the secondary consumers in the system (predators and omnivores) followed very similar patterns of mean abundance, showing a steady increase from spring to autumn.

These patterns suggest the possibility of a very intriguing interaction between nematode trophic levels in the system. Primary consumers increased in abundance from winter to spring for all feeding types. Similarly, predators also showed an increase in abundance during this same time frame. It is only during the transition from winter to
spring that this pattern was observed. The large standard error, coupled with the small sample size in winter (n=12), suggested that this difference may have been the result of sampling problems. However, the fact remains that during this time period, an increase in abundance was observed for both primary and secondary consumers, which suggests that an interaction was taking place between nematodes in these trophic levels. Table IV-1 shows that prey items, including bacterivorous nematodes, abounded in the system, and could conceivably support increasing numbers of predacious nematodes during this time period.

The primary predator genera in the current study included *Iotonchus* and *Prionchulus*, and the primary (and only) omnivorous genus in the system was *Dorylaimus*. The predator *Seinura* was also observed in the system, but in very low abundance. Khan and Kim (2007) concluded that all four of these genera are capable of preying on other nematodes and, in fact, do so as a primary means of nutrient acquisition. Additionally, Shafquat et al. (1987) showed that dorylaimid nematodes (in particular *Dorylaimus stagnalis*) prey upon nematodes of the genus *Cephalobus* (among others), which were found and classified as bacterivores in the current system (albeit in relatively low numbers). Furthermore, Shafquat et al. (1987) and Michiels and Traunspurger (2004) showed that as prey numbers increase, so too did the number of dorylaimid predators. A review of predatory nematodes by Small (1987) further supports these assertions. Therefore, it is reasonable to conclude that an increase in primary consumer abundance in spring contributed to an increase in secondary consumer abundance.

Further support for the interaction of these trophic groups was observed in both summer and autumn. Specifically, as the numbers of predators and omnivores increased
in summer, there appeared to be a concomitant decrease in primary consumer abundance. A suppressive impact on the abundance of prey nematodes would be expected as the number of predatory nematodes increased. Interestingly, a switch in the predator community structure (*Prionchulus* became the less dominant, and *Iotonchus* became the more dominant predator) also occurred in the transition from spring to summer. However, due to the generalist feeding habits of mononchid predators (Bilgrami et al. 1986), this switch would likely have little consequence on the suppressive ability of predacious nematodes on their nematode prey items. Furthermore, the most abundant bacterivore genus during this period was *Monhystera*, which is a common prey item for at least one of these genera (Table IV-1).

In autumn, primary consumers abundance appeared to rebound even as the number of secondary consumers increased. This trend was most obvious for bacterivores, which ended up dominating the nematode community during this season. Thus, there appeared to be no lasting negative implications of predators and omnivores preying on bacterivores in this system. There are several potential explanations for why such a trend might be observed. Yeates (1973) described that predatory nematodes are often much larger than bacterial feeding nematodes. Wardle et al. (1995) added that bacterivorous prey items are capable of supporting an initial increase in predator abundance; however, as predators continue to grow, the bacterivorous nematodes that were capable of sustaining predators when they were smaller, are no longer able to do so. Thus, Wardle et al. (1995) suggested that predatory nematodes switch to a different, larger food source, an ability that is made possible due to predator’s characteristic prey-generalist feeding strategy. This assertion is further supported by Yeates (1987), who concluded that
predatory nematodes often feed on different prey at different stages of their development. Predators tend to be resource limited (Yeates 1969, Wardle and Yeates 1993), and must adjust their prey capture accordingly to support further growth and development. Therefore, the increased abundance of both bacterivores and predators that was observed as the system transitioned from summer to autumn could have resulted from a switch in prey items by the predators from a smaller food source to a larger, more substantial and, therefore, more energy rich, one.

A second explanation could be that there were simply so many bacterivores present, that the bacterivorous population was not negatively impacted by the increased predator presence. Odum (1985) proposed that as succession progresses, as appeared to be occurring in the current system on an annual scale, larger organisms would be favored. Wardle et al. (1995) support this statement in their successional study in sawdust. As succession progressed, fungi began to prosper, improving the quality of the sawdust for the bacterivorous nematodes. Fungi supplemented the sawdust ecosystem by aiding in the breakdown of complex substrates (Lockwood 1981), which, consequently facilitated bacterial growth, i.e., food for the bacterivores, while also producing simpler compounds, which could be absorbed and utilized by the bacterivores. This is in accordance with Odum (1985) in that fungi tend to be larger than bacteria. This would then contribute to increased availability of resources to bacterivores, leading to larger bacterivores, which are capable of supporting a growing population of larger predatory nematodes (Wardle et al. 1995). The extent to which the current study adheres to this hypothesis is yet unclear because the abundance of bacteria and fungi were not measured. Furthermore, large amounts of disturbance are often observed in the littoral zones of freshwater lakes due to
factors like wind-induced wave action, and water-level fluctuations (Peters et al. 2007), which would suggest frequent stresses on the process of succession. As a result, it is perhaps less likely that the coordinated increase in both bacterivore and top predatory nematode abundances is due to this sequence of events. Rather, it seems most reasonable that the top predatory nematodes in the current system switched prey items, allowing the bacterivore community to increase simultaneously with the top predatory nematode (predators and omnivores) community.

There is some question concerning the effect of secondary consumers on the unicellular eukaryote feeders because as the numbers of predators and omnivores increased, the numbers of unicellular eukaryote feeders decreased. Two trends in the data make more confident assessment of this relationship difficult. First, the mean abundance of this feeding type was highest in spring, when the abundance of secondary consumers was still relatively low, and decreased the following season as predator and omnivore numbers increased (Fig. IV-4). Unicellular eukaryote feeders rebounded in abundance in autumn, but never attained the same level as they had in spring (though the difference in means between spring and autumn was not statistically significant). This trend is in accordance with the findings of Yeates (1987) and Wardle et al. (1995) in that it is likely that the top predatory nematodes switched to an alternate food source as they developed and matured. It is reasonable to assume that the unicellular eukaryote feeders were larger than bacterivorous nematodes based on the fact that the primary food source of the former is larger than that of the latter. As such, the resource-limited predators (Yeates 1969, Wardle and Yeates 1993) would require a more substantial food item than bacterivores alone to support their increased energetic demands. These data, therefore,
suggest that there was a negative impact on the unicellular eukaryote feeders in this system; however, it remains unclear as to whether or not it was the result of the predators and omnivores (top-down regulation) or some other factor that was not directly measured.

Second, the relationship between prevalence and season for unicellular eukaryotes was significant. This suggests that perhaps as the study progressed, unicellular eukaryote feeders dispersed more, thereby increasing the number of sites they inhabited while at the same time potentially decreasing the number of nematodes of this feeding type recovered in each sample.

**Influence of substratum type**

In general, it appeared that substratum type played a role in structuring the nematofaunal community composition of the system on a global scale. Bacterivores appeared to be least influenced by substratum type as they were the most prevalent feeding type observed in all substrata (Fig. IV-1). Additionally, they had the highest mean abundance of any feeding type for all substrata except in mixed substratum, where both bacterivores and predators were observed in nearly equal abundance (Fig. IV-4). Although bacterivores, omnivores, and predators all appeared to show the same general trends in prevalence, omnivores appeared to be the most selective of the substrata in which they were found. All three feeding types were more prevalent in mixed and mud/clay substrata, and less prevalent in leaf and mud/algae substrata (Fig. IV-2). The greatest dichotomy in this trend was observed for the omnivorous nematodes. This feeding type was much more prevalent in mixed and mud/clay substrata compared to leaf and mud/algae substrata. Perhaps the most logical explanation for the substratum specificity of this feeding type deals directly with its method of feeding. Linford and
Oliveira (1937) reported that dorylaimid predators depend solely on chance encounters to obtain prey. It is, therefore, reasonable to expect that, at least in mixed and mud/clay substrata compared to leaf substratum, the chances of encountering prey in a more open, i.e., less obstructed, habitat with fewer potential shelters for prey items, would be greatly increased. Based on this rationale, however, it remains unclear why omnivores were not more prevalent in the mud/clay substratum.

Omnivores were significantly more abundant in mixed substratum and significantly less abundant in leaf substratum than any other substrata (Fig. IV-6), implying that mixed was the most preferred substratum type and leaf the least preferred substratum type for this feeding type. Because mixed substratum contains elements of all substrata, it seems reasonable to find an omnivorous nematode genus in high abundance, as there would likely be multiple food (energy) sources present.

The community structure of predators and bacterivores observed in leaf and mixed substrata can be explained by the prey-generalist characteristic of mononchid predators. Specifically, the relative abundance of the bacterivore *Monhystera* was highest in leaf substratum where the prevalence of the predator *Prionchulus* was also high. Furthermore, in mixed substratum, the abundance of the predator *Iotonchus* was high, but the abundance of the bacterivorous *Monhystera* did not appear to be negatively impacted. The review by Small (1987) evidenced that *Monhystera* spp. are a common prey item for *Prionchulus* predators, and that a host of other primary–consumer nematodes are potential prey items for both of these genera. However, both, as are most mononchid predators, are prey-generalists (Small 1987, Bilgrami et al. 1986). Based on the inherent nature of both of these substrata, i.e., abundant leaf, stick, and organic cover,
it is likely that a large number of potential prey items were found there. Furthermore, the presence of organic matter would likely offer a good deal of shelter to these prey items. Thus, the presence of various prey items and increased shelter explains why an increase in predators may not result in a decrease in one particular prey item. However, the precise dynamics of these substrata were not explored in depth in the current study, so this explanation remains purely speculative.

*Monhystera* abundance was relatively low in mud/algae substratum where the predator *Iotonchus* was observed in greater abundance. These data suggest that this predator may have played a role in regulating the abundance of the prey *Monhystera*. Peters et al. (2005) point out that predation (top-down impact) likely directly influences the abundance of nematode genera that share a close association with periphyton. However, some questions remain to be answered concerning the substantiality of this predator-prey relationship as the composition of the periphyton (bottom-up impact) likely also directly influences the abundance of these organisms (Peters et al. 2005).

The fact that herbivores were both more prevalent (Fig. IV-2) and significantly more abundant in mud/clay substratum than any other substrata (Fig. IV-5) suggests that this was the preferred substratum type for these nematodes. This is logical in this system because the majority of the aquatic vegetation observed was present in this substratum type. The lack of a distinct littoral zone in most leaf transects, and decreased sunlight availability for the growth of aquatic vegetation because of leaf and other organic matter in mixed substratum transects, appeared to impact the amount of aquatic vegetation in these transects. *Lelenchus*, which is a root hair and epidermal cell feeder, was the only herbivore genus observed in this system and would, based on its life history, be expected
to inhabit those areas where aquatic vegetation is most abundant. This was indeed observed.

Finally, substratum type appeared to have the least impact on unicellular eukaryote feeders as there was no significant difference in prevalence (Fig. IV-2), and the only significant difference in mean abundance occurred between mixed and mud/algae substrata (Fig. IV-5 & IV-6). This feeding type was significantly more abundant in the former than the latter. This difference was surprising because meiofauna, e.g., diatoms and algae, both staples in the diet of unicellular eukaryote feeders, are typically associated with, or compose, the periphyton in the shallow littoral zones of lakes (Peters et al. 2007). The discontinuity and variability in the presence of large algal stands in the current study site may help explain this discrepancy.

Bilgrami AL, Ahmad AL, & Jairajpuri  1984.  Observations on the predatory behaviour 

morphological characters with molecular trees, and translating clades into ranks 

Ferris VR  1968.  Biometric analysis of the genus  labronema (Nematoda: Dorylaimida) 

Gomes GS, Huang SP, & Cares JE  2003.  Nematode community, trophic structure, and 
population fluctuation in soybean fields.  Fitopatol. Bras.  28:  258-266.

Hollis JP  1957.  Cultural studies with  Dorylaimus ettersbergensis.  Phytopathology  47:  
468-473.

Khan Z & Kim YH  2007.  A review on the role of predatory soil nematodes in the 

Linford MB & Oliveira JM  1937.  The feeding of hollow-spear nematodes on other 


Table IV-1: Predators observed in the current system and the prey items they consume. Listed are all nematode and non-nematode prey items observed in the current system (although others, not observed in Mallard Lake, may exist). (+) demarks those prey items consumed by a particular predator genus. Adapted from Small (1987).

<table>
<thead>
<tr>
<th>Order</th>
<th>Predator</th>
<th>Nematodes</th>
<th>Nematode Eggs</th>
<th>Protozoa</th>
<th>Cladocera</th>
<th>Rotifers</th>
<th>Oligochaetes</th>
<th>Agama</th>
<th>Diatoms</th>
<th>Bacteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dorylaimida</td>
<td>Enolaimida sp.</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Mononchida</td>
<td>Isochaeta sp.</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Mononchida</td>
<td>Lepeophtheirus sp.</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Mononchida</td>
<td>Monohysteria sp.</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Naidida</td>
<td>Frenchnema sp.</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Trichodorida</td>
<td>Trichodorus sp.</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Tylenchida</td>
<td>Syronema sp.</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

Adapted from Small (1987).
Table IV-2: Feeding type prevalence for the overall system, for each season, and for each substratum type. Columns represent the percent of the total number of sites in each sample containing at least one individual of the corresponding feeding type.
B=bacterivore, UEF=unicellular eukaryote feeder, H=herbivore, O=omnivore, and P=predator.

<table>
<thead>
<tr>
<th></th>
<th>Feeding Type</th>
<th>B</th>
<th>UEF</th>
<th>H</th>
<th>O</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overall</td>
<td></td>
<td>51.8</td>
<td>21.3</td>
<td>3.1</td>
<td>29.4</td>
<td>17.1</td>
</tr>
<tr>
<td>Season</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Winter</td>
<td></td>
<td>33.3</td>
<td>16.7</td>
<td>0.0</td>
<td>8.3</td>
<td>0.0</td>
</tr>
<tr>
<td>Spring</td>
<td></td>
<td>42.2</td>
<td>18.1</td>
<td>1.2</td>
<td>13.3</td>
<td>12.0</td>
</tr>
<tr>
<td>Summer</td>
<td></td>
<td>52.4</td>
<td>21.1</td>
<td>0.6</td>
<td>30.1</td>
<td>11.4</td>
</tr>
<tr>
<td>Autumn</td>
<td></td>
<td>61.5</td>
<td>25.0</td>
<td>9.4</td>
<td>44.8</td>
<td>33.3</td>
</tr>
<tr>
<td>Substratum Type</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leaf</td>
<td></td>
<td>51.1</td>
<td>22.2</td>
<td>1.1</td>
<td>13.3</td>
<td>11.1</td>
</tr>
<tr>
<td>Mixed</td>
<td></td>
<td>59.0</td>
<td>29.5</td>
<td>1.3</td>
<td>46.2</td>
<td>17.9</td>
</tr>
<tr>
<td>Mud/Algae</td>
<td></td>
<td>47.9</td>
<td>19.8</td>
<td>0.0</td>
<td>14.6</td>
<td>6.3</td>
</tr>
<tr>
<td>Mud/Clay</td>
<td></td>
<td>50.5</td>
<td>15.1</td>
<td>9.7</td>
<td>46.2</td>
<td>33.3</td>
</tr>
</tbody>
</table>
Fig. IV-1: Feeding type prevalence based on season.
### Seasonal Prevalence of Feeding Strategies

<table>
<thead>
<tr>
<th>Season</th>
<th>Prevalence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Winter</td>
<td>10.0</td>
</tr>
<tr>
<td>Spring</td>
<td>20.0</td>
</tr>
<tr>
<td>Summer</td>
<td>30.0</td>
</tr>
<tr>
<td>Autumn</td>
<td>40.0</td>
</tr>
</tbody>
</table>

**Legend:**
- Bacterivore
- Unicellular Eukaryote Feeder
- Herbivore
- Omnivore
- Predator
**Fig. IV-2:** Feeding type prevalence based on substratum type.
Fig. IV-3: Mean abundance based on feeding type. Error bars are standard error. Bars with the same letter are not significantly different from one another.
Mean Nematode Abundance

Feeding Type

- Bacterivore
- Unicellular Eukaryote Feeder
- Herbivore
- Omnivore
- Predator

Different letters indicate significant differences among feeding types.
**Table IV-3:** Genera represented in each feeding type observed. Values are expressed in the total number of individuals observed belonging to each genus (n), the total number of individuals within each feeding type (total), and the relative abundance of each feeding type (%).

<table>
<thead>
<tr>
<th>Feeding Type</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Bacterivore</strong></td>
<td></td>
</tr>
<tr>
<td>Monhystera</td>
<td>633</td>
</tr>
<tr>
<td>Monochromadora</td>
<td>92</td>
</tr>
<tr>
<td>Terschellingia</td>
<td>60</td>
</tr>
<tr>
<td>Thalasomonhystera</td>
<td>23</td>
</tr>
<tr>
<td>Bunomhystera</td>
<td>22</td>
</tr>
<tr>
<td>Prisnatoicae</td>
<td>19</td>
</tr>
<tr>
<td>Rhadidolaimus</td>
<td>17</td>
</tr>
<tr>
<td>Anoplodrillus</td>
<td>16</td>
</tr>
<tr>
<td>Panagrolaimus</td>
<td>11</td>
</tr>
<tr>
<td>Cionechus</td>
<td>10</td>
</tr>
<tr>
<td>Chromogaster</td>
<td>9</td>
</tr>
<tr>
<td>Ocholaimus</td>
<td>8</td>
</tr>
<tr>
<td>Plectus</td>
<td>8</td>
</tr>
<tr>
<td>Paraplectonema</td>
<td>6</td>
</tr>
<tr>
<td>Monhystrella</td>
<td>3</td>
</tr>
<tr>
<td>Alaimus</td>
<td>2</td>
</tr>
<tr>
<td>Rhadidoides</td>
<td>2</td>
</tr>
<tr>
<td>Faraphanolaimus</td>
<td>1</td>
</tr>
<tr>
<td>Cephalobus</td>
<td>1</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>993 (51.8%)</td>
</tr>
<tr>
<td><strong>Predator</strong></td>
<td></td>
</tr>
<tr>
<td>Ictonchus</td>
<td>310</td>
</tr>
<tr>
<td>Prionchulus</td>
<td>86</td>
</tr>
<tr>
<td>Ironus</td>
<td>3</td>
</tr>
<tr>
<td>Tripyla</td>
<td>2</td>
</tr>
<tr>
<td>Faramononchus</td>
<td>1</td>
</tr>
<tr>
<td>Steinsura</td>
<td>1</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>409 (21.3%)</td>
</tr>
<tr>
<td><strong>Omnivore</strong></td>
<td></td>
</tr>
<tr>
<td>Dolcholaimus</td>
<td>273</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>273 (14.2%)</td>
</tr>
<tr>
<td><strong>Unicellular Eukaryote Feeder</strong></td>
<td></td>
</tr>
<tr>
<td>Sertobrillus</td>
<td>111</td>
</tr>
<tr>
<td>Neobrillus</td>
<td>106</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>217 (11.3%)</td>
</tr>
<tr>
<td><strong>Herivore</strong></td>
<td></td>
</tr>
<tr>
<td>Lelenciulus</td>
<td>26</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>26 (1.3%)</td>
</tr>
</tbody>
</table>
**Fig. IV-4:** Mean abundance of nematode feeding types based on season. Error bars are standard error.
**Fig. IV-5:** Mean abundance of each feeding type based on substratum type. Error bars are standard error.
Fig. IV-6: Mean nematode abundance in each substratum type based on feeding type. Error bars are standard error.
CHAPTER V

The Role of Spatiotemporal Influences on Prevalence, Mean Abundance, and the Overall Community Dynamics of Free-living Nematodes Classified by Feeding Method in a Freshwater System

ABSTRACT

Few studies have attempted to better understand the influence of abiotic and biotic factors on the composition of the free-living nematode communities in freshwater systems. The current study was undertaken to discern the role of spatiotemporal factors, e.g., season and substratum type, on the community composition of the free-living nematofauna in a freshwater system. Substratum samples were collected from January through October, 2009 from Mallard Lake (a 4.9 ha, eutrophic pond located in the Piedmont region of North Carolina). Nematodes were recovered from substratum samples via the Baermann extraction method, isolated, identified, and quantified. In an attempt to achieve finer resolution of the observed fluctuations in community composition in the system, all identified nematodes were classified into 4 groups: (1.) deposit feeders/swallowers, (2.) epistrate feeders, (3.) chewers, and (4.) suction feeders. For the purposes of the current study, a fifth feeding method (parasitic) was added. Prevalence and mean abundance values were calculated based on feeding method for: (1.) the entire system, (2.) each season, and (3.) each substratum type observed.
A total of 1,928 nematodes was recovered, representing 44 species and 30 genera. Deposit feeders accounted for 44.8% of all nematodes recovered, and the system was dominated by the genus *Monhystera*. All parasites observed in the system were keyed to the order Mermithida. Season, trophic interactions, and substratum type all influenced the community composition of nematodes utilizing various feeding methods in the system. Deposit feeder and suction feeder prevalence was most influenced by seasons, and mean abundance of all feeding methods, with the exception of deposit feeders, was also influenced by this variable. A complex trophic interaction was observed in which the nematode community appeared to switch from a bottom-up regulated system early in the study, to a top-down regulated system later in the study. Substratum type appeared to have the greatest impact on the prevalence and mean abundance of suction feeders and chewers. Deposit feeders were dominant in nearly every season and every substratum type, and showed little fluctuation in both prevalence and mean abundance.
INTRODUCTION

Relatively few studies exist that attempt to explain the influence of spatiotemporal factors on the community composition of the free-living nematofauna in freshwater systems. Still fewer studies attempt to do so in a eutrophic system. Those that take this approach appear to be mixed in their conclusions. On the one hand, several studies suggest that both spatial (Prejs 1977, Vidaković and Bogut 2004, Bert et al. 2007) and temporal factors (Vidaković and Bogut 2004) influence free-living nematode assemblages. Conversely, Michiels and Traunspurger (2004) reported that seasonality had very little influence on the free-living nematode community in a eutrophic lake, Lake Obersee, in Germany. The present study provides a rare look at the role of both of these factors in a eutrophic ecosystem. Studies that explore the influence of both spatial and temporal variables on the community structure of free-living nematodes are capable of providing answers to questions that could not be attained by looking at either factor alone (Gomes et al. 2003).

Here, nematode prevalence and mean abundance were used as measures of the influence of season and substratum type on these meiobenthic organisms. Nematodes were collected from the substratum, recovered via Baermann extraction techniques, isolated, identified to genus, and quantified. Nematodes were then placed into groups based on buccal morphology as described by Traunspurger (1997). Four classifications were utilized to group the nematodes in this system: (1.) deposit feeder/swallower, (2.) chewer, (3.) epistrate feeder, and (4.) suction feeder. Fig. V-1 provides pictorial examples of the buccal morphology associated with each of these feeding methods. A
fifth group, parasitic nematodes, was added to the classification system proposed by Traunspurger (1997) for the purposes of the present study.

Traunspurger (1997) offers an in-depth description of the buccal morphology associated with each of the feeding methods he described. To summarize, deposit feeder/swallowers typically lack teeth. Wieser (1953) added that, depending on whether these individuals are selective or non-selective deposit feeders, the size of the buccal cavity will vary. Specifically, the former has a smaller cavity that allows only bacterial sized particles to enter, while the latter possesses a more spacious buccal cavity (Wieser 1953). The present study did not subdivide the observed deposit feeders into groups on this basis. Traunspurger (1997) explains that epistrate feeders possess only a small tooth used to break, or tear, food particles apart as they enter the buccal cavity. Chewers have voluminous, highly sclerotized buccal cavities containing one or more teeth, denticles, or both. Suction feeders bear a stylet, which is used to pierce food items and suck out contents.

Traunspurger (1997) also provides a brief description of the food items that each of these feeding methods typically consume, as well a description of the manner in which some of them feed. Deposit feeders typically feed on bacteria and unicellular eukaryotes, and do so by swallowing their food whole. Epistrate feeders, i.e., tear-and-swallow feeders, utilize their small tooth to feed on bacteria, unicellular eukaryotes, diatoms, and other microalgae. Chewers usually feed on a number of larger meio-benthos, e.g., unicellular eukaryotes, nematodes, rotifers, tardigrades, and enchystraeids, and do so by breaking these food items into smaller bits with their teeth and denticles. Suction feeders
are typically classified as omnivores and feed on an array of food items, e.g., algae, plants, fungi, and animals.

The purpose of the present study was multifaceted. First, an understanding of the contribution of nematodes utilizing various feeding methods to the overall nematode community in the freshwater system was desired. Second, an effort was made to understand the nature of spatiotemporal factors in driving the patterns of prevalence and abundance of nematodes possessing different buccal armament, i.e., utilizing the 5 different feeding methods listed above. Finally, the study was interested in understanding the trophic relationships that existed between these different groups of nematodes.
MATERIALS AND METHODS

Sampling method

Samples were collected from Mallard Lake according to the methods previously described (Chapter III).

Mo were grouped into seasons as described in Chapter IV.

Nematode recovery

Nematodes were recovered using a Baermann-funnel apparatus, identified, and stored as described in Chapter III.

Feeding type classification

Nematodes were placed into groups based on buccal morphology as described by Traunspurger (1997). All morphotyped nematodes were classified as (1.) deposit feeders/swallowers, (2.) epistrate feeders, (3.) chewers, (4.) suction feeders, or (5.) parasitic. Several key morphological features pertaining specifically to the buccal region of the nematodes (the extent of buccal sclerotization; the presence or absence, number, and size of teeth, denticles, or both; and the presence or absence of an odontostyle or stomatostyle) were considered when classifying nematodes into the first 4 feeding-method groups. The use of buccal morphology to make predictions about feeding behavior in nematodes is quite common, and has been used in a number of systems, including both marine (Wieser 1953) and freshwater (Traunspurger 1997; Moens et al. 2004). A fifth group (parasitic) was added to this classification system for the purposes of the present study because this group was of particular interest to the author.

Morphological descriptions of the mermithids, here classified as parasitic, remains scant
and definitive grouping into one of the four groups described by Traunspurger (1997) was not possible. They were thus considered as a separate group.

Data analysis

Of the 2,349 total nematodes collected, only 1,828 were identified to morphotype. This discrepancy was the result of 1 of 3 factors: (1.) morphotyping did not begin until the 26 March 2009 collection so the identity of any nematodes collected prior to this date is unknown, (2.) some nematodes were too small to be clearly identified, or (3.) a very small number of nematodes were lost during transfer from the stereomicroscope to the bright field microscope.

Because individual identification of nematodes did not begin until the final sampling event in March 2009, the data set for this mo (and consequently winter as a whole) consisted of only 12 independent samples. This oversight is mentioned here in the spirit of full disclosure, and was taken into consideration when analyzing these data. Feeding-method prevalence and mean abundance values were calculated both seasonally and based on substratum type. Multiple regression analysis was used to determine whether a significant relationship existed between prevalence and season across all feeding methods, and individual regression analyses were performed to determine whether the relationship between prevalence and season for each individual feeding method was significant.

Two series of Chi-square analyses were conducted on the feeding method data based on season and substratum type. The first looked at each season or substratum type separately, and sought to determine whether significant differences existed between observed and expected prevalence values for all 5 feeding methods, i.e., data classes.
The observed numbers for each data class were simply the number of times each feeding method was observed, i.e., presence, in each season or substratum type. Expected values for these analyses were generated by multiplying the sum of the observed presence values for all feeding methods, times the proportion of each feeding method actually observed in the system as a whole, i.e., the prevalence of each feeding method (see Chapter III). Because the percent contribution of each feeding method to the overall abundance observed in the system was known, these proportions could be used to generate the expected values for each data class, i.e., the number of times one would expect to observe each feeding method in each season or substratum type based on what was actually observed in the system.

The second series of Chi-square analyses sought to determine whether observed prevalence values differed significantly from expected values between seasons or substrata for each feeding method. Therefore, each feeding method was analyzed separately, and the data classes considered in these analyses were either seasons or substrata. Observed values for each data class corresponded to the observed presence data for said feeding method in each season or substratum type. The expected proportions utilized in these analyses were based on the proportions of all the nematodes in the system found in each season or substratum type, i.e., the calculated nematode prevalence obtained for each season or substratum type in the system as a whole (see Chapter III). The expected number for each data class, i.e., season or substratum type, was generated by multiplying the summed seasonal or substrata presence values for each feeding method, and the proportion of nematodes found in each season or substratum type for the whole system (see Chapter III). Further discussion of trends for both series
of Chi-square analyses, i.e., determining which feeding methods, seasons, or substrata deviated the most from expected values, was purely observational, and was not analyzed statistically due to the subsequent loss of statistical power associated with conducting multiple Chi-square analyses with low degrees of freedom.

Data collected for the current study consistently failed the $F_{\text{max}}$ test for equal variance. To decrease the chance of committing type II (false negative) errors, these data were not analyzed with standard parametric statistical tests, e.g., ANOVA. Ordinarily, when the assumptions of normality and equal variance are not satisfied, a non-parametric analog, e.g., the Kruskal-Wallis test, can be employed. However, the use of these tests incur limitations in there own right, including most notably, a loss of statistical power when compared to parametric tests. Therefore, the current study utilized Monte Carlo randomizations to determine the significance of differences between means within and among samples.

This method of analysis offers a powerful alternative to parametric tests because it randomly re-samples the collected data set, thereby removing the constraints of normality and equal variance encountered when using parametric tests. Specifically, the difference in mean abundance values for every possible pairwise combination of seasons or substratum type was calculated for each feeding method separately. The data set was then randomly rearranged 30,000 times, and the pairwise differences in mean abundance between each season or substratum type were re-calculated from the randomized data set. The calculated pairwise differences resulting from the 30,000 random iterations were then compared to the actual differences in mean abundance observed between seasons or substratum type in the system, i.e., the actual observed mean abundance differences, prior
to random rearrangement. P-values were generated by dividing the number of times the randomized mean difference exceeded or was equal to the actual observed mean difference between seasons or substrata, by the total number of iterations. When the iterative mean abundance differences were greater than or equal to the actual mean abundance differences more than 5% of the time, the differences in actual observed means were not considered significant. The same series of analyses were performed to compare the difference in mean abundance between every pairwise combination of feeding methods for every season and substratum type. The significance threshold was set at $p \leq 0.05$ for all analyses.

**Terminology**

The term relative abundance is defined in the context of the current study as the percent of the total nematofauna contributed by any 1 feeding method or genus. The use of the terms prevalence, mean abundance, sampling units, morphotypes, and genus names were described in Chapter III.
RESULTS

Overall nematode assemblage based on feeding type

A total of 1,928 nematodes was classified into the 4 feeding methods proposed by Traunspurger (1997), or as parasitic. Individuals from 30 genera were observed. Additionally, 8 individuals were identified to the order Mermithida. Of the 30 reported genera, the 5 most commonly observed (Monhystera, Iotonchus, Dorylaimus, Semitobrilus, and Neotobrilus) contributed 77.2% of the total individuals recovered. Nearly half (44.8%) of the nematode community was composed of individuals classified as deposit feeders, or swallowers (Table V-1), which ingest and swallow their food whole (Traunspurger 1997). Nematodes utilizing this feeding method were both the most prevalent (Table V-2) and the most abundant observed (Fig. V-4). Deposit feeders were identified in more samples (48.7%) and in significantly greater numbers than nematodes employing any of the other feeding methods (Appendix B-4). Nematodes from 15 genera and 20 morphotypes possessed buccal morphology supporting their classification within this group, making it the most common feeding method as well (Table V-1). More than 75% of the individuals utilizing this method of nutrient acquisition came from the genus Monhystera, 7% belonged to Terschellingia, and the remaining 18% came from the other 13 genera.

Chewing type feeders were the second most commonly observed, comprising 34.6% of the total nematode community (Table V-1). Chewers were observed in 30.9% of samples (Table V-2) and were significantly less abundant than deposit feeders, but significantly more abundant than epistrate feeding, suction feeding, and parasitic
nematodes (Fig. V-4). *Iotonchus* contributed 50% of these individuals while *Semitobrilus*, *Neotobrilus*, and *Prionchulus* contributed 17.6, 16.8, and 13.6% of these individuals, respectively. The remaining genera contributed relatively few nematodes. Suction feeders, accounting for 15.6% of the total nematodes observed (Table V-1), were recovered from 31.2% of samples (Table V-2), and were significantly more abundant than either epistrate feeders or parasites (Fig. V-4). *Dorylaimus*, the dominant suction feeding genus, contributed 91.3% of the individuals using this feeding method, with *Lelenchus* contributing the rest.

Epistrate feeders, with a relative abundance of 6.4%, were the fourth most abundant (Fig. V-4) and the fourth most prevalent feeding method observed (Table V-2). The epistrate-feeding nematodes were predominately *Monochromadora*, which alone, comprised 74.2% of the nematodes employing this feeding method. Finally, the least common feeding method observed was parasitism, which accounted for 0.4% of the total nematode community (Table V-1) and was observed in only 1.9% of all samples (Table V-2). All parasitic nematodes observed in the study were classified as belonging to the order Mermithida.

**Prevalence based on feeding method**

*Season:* There was no significant relationship between prevalence and season when all feeding methods were considered together ($R^2=0.833$, $p=0.087$). Presence values differed significantly from expected values during spring, summer, and autumn ($\chi^2$ test, $p=0.052$, $p<0.001$, and $p<0.001$, respectively). During spring, chewers were present in fewer sites than expected, while epistrate feeders were recovered from more sites than expected. In summer, chewers were again recovered from a fewer number of sites than
expected, while both suction feeders and epistrate feeders had presence values greater than expected. In autumn, suction feeders were again observed in a greater number of samples than expected, as were parasites. Observed presence values for all feeding methods were not significantly different than expected in winter ($\chi^2$ test, $p=0.937$).

Prevalence values of deposit feeders were consistently higher than any of the other feeding methods observed (Fig. V-2), and were significantly influenced by season ($R^2=0.931$, $p=0.035$). Furthermore, seasonal observed presence of nematodes in this group differed significantly from those expected, based on observed seasonal nematode prevalence in the system as a whole ($\chi^2$ test, $p<0.001$). The greatest amount of deviation for deposit feeders occurred in winter, when they were recovered from 80% fewer sites than expected.

Chewing and suction feeding nematodes also showed positive trends in prevalence across seasons. This relationship was not significant for chewers ($R^2=0.887$, $p=0.058$), but was significant for suction feeders ($R^2=0.963$, $p=0.019$). Furthermore, the observed prevalence values for both chewing and suction feeding nematodes were significantly different than expected between seasons ($\chi^2$ test, $p=0.001$ and $\chi^2$ test, $p<0.001$, respectively). For both feeding methods, observed presence values were less in winter and greater in autumn than expected.

A similar trend was observed for epistrate feeders in that observed presence differed significantly from expected values on the basis of season ($\chi^2$ test, $p=0.034$); however, there was not a clear positive trend in prevalence across seasons for this feeding method like was observed for the other two feeding methods. Epistrate feeders were consistently less prevalent than deposit feeders, chewers, and suction feeders with the
exception of the winter mo, where they occurred with the same prevalence (9.1%) as suction feeders (Fig. V-2). Furthermore, observed presence was lower in winter and higher in spring than expected for this feeding method. Parasites were consistently less prevalent than all other feeding strategies observed for all seasons. There was no significant relationship between prevalence and season for this feeding method ($R^2=0.772$, $p=0.121$), and the observed presence values did not differ significantly from those expected on the basis of season ($\chi^2$ test, $p=0.009$).

*Substratum type:* Presence values differed significantly from those expected within feeding methods in mixed, mud/algae, and mud/clay substrata ($\chi^2$ test, $p<0.001$, $p<0.001$, $p<0.001$, respectively). Presence was not significantly different from expected in the leaf substratum ($\chi^2$ test, $p=0.827$). Deposit feeders were the most prevalent nematodes in all substrata, although in mud/algae substratum they occurred in nearly the same number of samples as suction feeders (Table V-2, Fig. V-3). Observed presence between substratum types did not differ significantly for this feeding type ($\chi^2$ test, $p=0.190$).

Chewers were more prevalent in mixed substratum and less prevalent in mud/clay substratum than the other two substrata (Fig. V-3). Presence values were significantly different from expected values on the basis of substrata ($\chi^2$ test, $p=0.004$). In particular, chewers were recovered more frequently in leaf and mixed substratum, and less frequently from mud/algae and mud/clay substrata than expected. Furthermore, chewers were observed in fewer sites than expected in both mud/algae and mud/clay substrata.

Suction feeders were most prevalent in mixed, mud/algae, and mud/clay substrata, and less prevalent in the leaf substratum (Table V-2, Fig. V-3). As with chewers, the differences in observed and expected presence values between substrata were significant.
The greatest deviations from expected values for this feeding method were observed in leaf, mixed, and mud/algae substrata. Specifically, observed presence in the former was lower than expected, and higher than expected in the remaining 2 substrata. Furthermore, suction feeders were observed in more samples than expected in mixed, mud/algae, and mud/clay substrata.

Prevalence of epistrate feeders was quite consistent across substrata (Fig. V-3), and observed presence values among substrata did not differ significantly from expected (χ² test, p=0.752). A slight peak in abundance was observed in the mud/algae substratum, which corresponded to large discrepancies in presence in mud/algae and mud/clay substrata when compared to expected presence. In both cases, epistrate feeders were observed in more sites than expected. Finally, the prevalence of nematodes classified as parasitic showed a peak in the mixed substratum, although the differences between observed and expected presence values among substrata were not significant (χ² test, p=0.155). This spike in prevalence corresponded to parasites being found in more sites than expected in the mixed substratum.

There appeared to be two clear patterns in the composition of the nematode community in this system based on substratum type. First, in leaf and mixed substrata, deposit feeders were the most prevalent, followed by chewers, suction feeders, epistrate feeders, and finally parasites. Although the same decreasing pattern was observed in both substrata, the prevalence of suction feeders was much higher and, therefore, closer to that of chewers in the mixed substratum. All other feeding methods showed remarkable consistency between these two substrata (Table V-2). Second, in mud/algae and mud/clay substrata, the prevalence of deposit feeders was again highest, but in this
case, suction feeders exhibited the second highest prevalence, followed by chewers, epistrate feeders, and finally parasites. Again, suction feeder prevalence was the only one to show much variability and was higher and, therefore, closer to that of deposit feeders in the mud/algae substratum.

**Abundance based on feeding method**

*Season:* Overall, a significant relationship between mean abundance and season was not observed ($R^2=0.013, p=0.887$). However, there did appear to be a positive trend in mean abundance for deposit feeders, chewers, and suction feeders (Fig. V-5). The relationship between mean abundance and season was significant for chewing nematodes ($R^2=0.943, p=0.029$), but not for deposit or suction feeders ($R^2=0.835, p=0.086$ and $R^2=0.742, p=0.139$, respectively). The mean abundance of deposit feeders did not differ significantly between seasons (Appendix B-6), but did differ significantly for chewers between autumn and summer when they were more abundant in the former than the latter. However, no other significant differences in mean abundance were observed for chewing nematodes between the remaining seasons (Appendix B-6).

Suction feeders showed the most variability of the three feeding types (Fig. V-5). Mean abundance values for nematodes utilizing this feeding strategy were significantly higher in summer than in spring, and significantly higher in autumn than in any other season (Appendix B-6). Epistrate feeders peaked in abundance in spring, but the difference was only significant between spring and autumn, and not spring and winter (Appendix B-6). Mean abundance of nematodes utilizing a parasitic feeding strategy showed an overall positive trend over the course of the study, although the relationship between mean abundance and season was not significant ($R^2=0.748, p=0.135$). Even so,
mean abundance was significantly higher in autumn than in winter and spring (parasites were absent in both of these seasons) (Fig. V-5, Appendix B-6).

Within season mean abundance differed significantly between feeding methods in all seasons (Appendix B-5). In winter, deposit feeders were significantly more abundant than chewers, epistrate feeders, and parasites. No other significant differences were observed between any of the other feeding methods (Appendix B-5). During spring, summer, and autumn, deposit feeders were significantly more abundant than epistrate feeders, parasites, and suction feeders. Chewers were significantly more abundant than parasites in spring, summer, and autumn; but were only significantly more abundant than suction feeders in spring and summer, and significantly more abundant than epistrate feeders in summer and autumn (Appendix B-5). Finally, the increase in mean abundance of suction feeders observed from summer to autumn made them significantly more abundant than epistrate feeders and parasites in autumn (Fig. V-5, Appendix B-5).

Two genera (*Terschellingia* and *Rhabdoalimus*) accounted for 58.8 and 23.5% (respectively) of the deposit feeders observed in winter, and together made up ≈50% of all nematodes recovered in this season. Individuals of *Dorylainus* (suction feeders) were the only other nematodes to appear with notable abundance, contributing 21.4% of the total nematodes observed in this season. No parasites were recovered in winter or spring (Table V-2, Fig. V-5). The onset of spring resulted in a clear shift in the deposit feeder community structure such that *Terschellingia* dropped from a relative abundance of 58.8% to 15.1%, while *Monhystera* increased from a relative abundance of 5.9% in winter to 53.6% in spring. This particular deposit-feeding genus dominated every season, reaching relative abundances of 81.9 and 96.6% in summer and autumn, respectively.
After spring, *Monhystera* was the only deposit feeding genus to contribute >5% of the relative abundance of nematodes using this feeding strategy. Nematodes of this genus, and therefore, the deposit feeding method itself, dominated all other genera and all other feeding methods throughout the remainder of the study.

A transition in the community composition of chewing nematodes was also observed over the course of the study. *Semitobrilus* was the dominant chewing nematode observed in both winter and spring (100 and 60.5%, respectively), but after the appearance of *Prionchulus* and *Neotobrilus* in the spring, the relative abundance of *Semitobrilus* consistently declined to the point where no individuals were recovered in August. The relative abundance of *Neotobrilus* increased consistently throughout the remainder of the study, showing a nearly 2-fold increase from one season to the next. In spring, summer, and autumn, this genus was observed with a relative abundance of 7.9, 12.1, and 28.0%, respectively. The chewer *Iotonchus* appeared as the dominant genus within this feeding method in the summer, reaching a relative abundance of 67.8%, and continued to dominate this feeding method into autumn (57.3%). The epistrate feeder *Monochromadora* was observed throughout all seasons and experienced relative abundances ranging from 41.7 to 73.3%, making it one of the most consistent epistrate feeders, and genera, observed. Individuals employing this feeding method dominated spring and summer (73.3 and 61.3%, respectively), but were relatively less abundant than *Prismatolaimus* in autumn. The relative abundance of *Prismatolaimus* increased consistently from spring to autumn, showing a nearly 2-fold increase in relative abundance between seasons. The relative abundance of *Prismatolaimus* increased from 11.1 to 22.6% from spring to summer, and from 22.6 to 58.3% from summer to autumn.
Dorylaimus had the highest relative abundance of suction feeding nematodes during all seasons (ranging from 62.5 to 100%). The only other suction-feeding genus (Lelenchus) was observed only in spring and autumn, and reached relative abundances of 25.0 and 13.8%, respectively. Finally, the parasitic specimens observed belonged to the order Mermithida and this feeding strategy was consistently less abundant than all others in any season (Fig. V-5).

**Substratum type:** Deposit-feeding nematodes were significantly more abundant than epistrate feeders and parasitic nematodes in all substrata, and were significantly more abundant than suction feeders and chewing nematodes in all but the mixed substratum (Fig. V-6, Appendix B-7). The mean abundance of deposit feeding nematodes was consistently high in all substrata, but did not differ significantly among substrata (Appendix B-8). Chewing nematodes were the only nematodes to outnumber deposit feeders in any substrata, although the differences in mean abundance in mixed substratum, where this occurred, were not significant (Fig. V-6, Appendix B-7). Chewing nematodes were significantly more abundant than parasites in all substrata, and were significantly more abundant than epistrate feeders and suction feeders in leaf and mixed substrata, but not mud/algae and mud/clay substrata (Appendix B-7). Even though the differences were not significant in mud/algae substratum, chewers still outnumbered both epistrate feeders and suction feeders (Fig. V-6). In mud/clay substratum, chewing nematodes outnumbered epistrate feeders, but were less abundant than suction feeders; however, these differences were not significant (Appendix B-7). Unlike deposit feeders, chewing nematodes showed significant variation in mean abundance among substrata and were
significantly more abundant in mixed substratum than in any other substrata. Their mean abundance was not significantly different in the remaining substrata (Appendix B-8).

Suction feeders were significantly more abundant than parasitic nematodes in both mud/algae and mud/clay substrata; however, they were significantly more abundant than epistrate feeders in only the mud/clay substratum (Appendix B-7). Suction feeders were more abundant than chewing nematodes in mud/clay substratum, but again this difference was not statistically significant (Fig. V-6, Appendix B-7). Suction feeders were significantly less abundant in leaf substratum than in any of the other substrata, and were significantly more abundant in mixed substratum than mud/clay substratum (Appendix B-8). Mean abundance of epistrate feeders and parasitic nematodes did not differ significantly in any substrata, although epistrate feeders were more numerous than parasitic nematodes in all substrata (Fig. V-6). There were no statistically significant differences in mean abundance values between substrata for either of these feeding methods (Appendix B-8).

Monhystera was the most abundant deposit-feeding group observed in all substrata. The relative abundance of this genus ranged from 73.8 to 82.7%, and only two other genera contributed greater than 5% of the mean abundance of this feeding type in any substrata. Thalassomonhystera accounted for 5.3% of the relative deposit feeder abundance in leaf substratum, while Terschellingia contributed 13.1% of the deposit feeders in mud/algae substratum. Monhystera accounted for 47.4, 34.4, and 43.2% of the relative abundance of all nematodes in leaf, mud/algae, and mud/clay substrata, respectively.
The chewing-feeding method was dominated by Neotobrilus, Iotonchus, Prionchulus, and Semitobrilus. There was much more variability in the dominant genus in each substratum type for chewers, though Iotonchus had the highest relative abundance in both mixed and mud/algae substrata (72.7 and 37.8%, respectively). Interestingly, this genus had the highest relative abundance of any nematode genus in mixed substratum (35.0%), but was almost entirely missing from mud/clay substratum. Although it was found in all substrata, Neotobrilus showed highly variable relative abundance as well, ranging from a low of 10.8% in mud/algae substratum to 30.3% in leaf substratum, where it was more abundant than any of the other chewing genera. Semitobrilus had the highest relative abundance within this feeding type in mud/clay substratum (56.7%), but was consistently less abundant relative to the other chewing genera in leaf, mud/algae, and mud/clay substrata (18.3, 20.7, and 5.2%, respectively). Prionchulus had consistently lower relative abundance than the other chewing genera with the exception of mud/algae substratum, where it had a relative abundance of 24.3% and was more abundant than both Semitobrilus and Neotobrilus.

Two epistrate feeding genera, Monochromadora and Prismatolaimus, dominated this feeding method. The relative abundance of Monochromadora was consistently higher than other epistrate feeding genera in leaf, mixed, and mud/algae substrata, while Prismatolaimus was relatively more abundant than the former in mud/clay substratum. Other notable genera utilizing this feeding method included Monhystrella, Oncholaimus, Ironus, and Tripyla.

Dorylaimus dominated the suction feeding method in all substrata, although Lelenchus and Seinura were also observed. In both mud/algae and mud/clay substrata,
*Dorylaimus* was the second most abundant nematode genus, relative to all other genera and feeding methods, contributing 17.3 and 19.2% of the total nematodes observed. All parasitic nematodes were classified in the order Mermithida and contributed less than 1% of the relative abundance among all nematode-feeding methods in all substrata.
DISCUSSION

**Analysis of the system based on feeding method**

Deposit feeders were the most prevalent nematodes observed in the system, followed by suction feeders, chewers, epistrate feeders, and finally parasites (Table V-2). Mean abundance of feeding methods followed a similar trend (Fig. V-4); however, chewing nematodes occurred in significantly higher number than suction feeders, and the mean abundances of epistrate feeders and parasites were not significantly different (Appendix B-4). Observed results were similar to those described in both oligo-mesotrophic and eutrophic lakes (Table 6.1 in Moens et al. 2006), and for glacier-fed streams (Eisendel 2008).

*Monhystera* was the dominant deposit feeder in this system and accounted for over 75% of the individuals observed. Although this genus is typically associated with oligotrophic systems (Strayer 1985, Wu and Liang 1999), Prejs (1977) reported finding it in both oligotrophic and eutrophic lakes, albeit typically in lower abundance in the latter than the former. Furthermore, Eisendle (2008) reported that *Monhystera* dominated freshwater glacial streams, a habitat that is usually considered to be extremely harsh. On the other hand, Monhysteridae, of which this genus is a member, has been reported to dominate other eutrophic systems, including a series of small farm ponds (Bert et al. 2007), and a few eutrophic lakes (Prejs 1977, Michiels and Traunspurger 2004).

The dominant chewer was *Iotonchus*, which accounted for 50% of the total chewing nematodes observed. *Dorylaimus* made up 91.3% of the suction feeding nematodes observed, while 74.2% of the epistrate feeders were composed of nematodes
belonging to *Monochromadora*. Finally, all parasites in the system belonged to a single order (Mermithida).

**Influence of season and trophic interactions**

Season played a role in structuring the nematode community when considering the feeding method these organisms utilize to obtain their food. No significant relationship existed for the system when all feeding methods were considered together. This is most likely due, at least in part, to the decrease in prevalence experienced by epistrate feeders in summer and autumn. With the exception of deposit feeders and epistrate feeders, significant relationships existed between prevalence and season. Furthermore, chewers, suction feeders, and parasites all showed significant increases in mean abundance between winter and autumn, suggesting that season may have the greatest influence on nematodes employing these feeding methods.

Clear differences in both prevalence and mean abundance among feeding methods were observed within each season (Fig. V-2 & V-5, respectively). Furthermore, when feeding methods were considered individually, significant differences in prevalence and mean abundance (Appendix B-6) were again observed. The influence of season was evident on both a large scale, i.e., by driving patterns concerning feeding methods, as well as on a smaller scale, i.e., by driving patterns concerning genera.

The relationship between prevalence and season was significant for both deposit feeders and suction feeders, suggesting that as the ecosystem moved from one season to the next, nematodes employing these feeding methods dispersed and were, therefore, observed in a greater number of samples with each successive season. Free-living nematodes are capable of both passive (Hagerman and Rieger 1981, Palmer 1988, Atilla
and Fleeger 2000) and active (Ullberg and Ólafsson 2003) dispersal. Because free-living nematodes are often located in the top 2 cm of sediment (Traunspurger and Drews 1996), and the water-substrate interface is associated with greater levels of erosion (Warwick and Gee 1984, Eskin and Palmer 1985, Palmer and Gust 1985), it is logical that as time progressed, and nematode abundance increased (Fig. V-5), more and more nematodes in the system dispersed (actively by crawling) or were dispersed (passively in the water column), thereby increasing nematode prevalence as well (Fig. V-2).

Significant differences in seasonal observed versus expected presence values for deposit feeders, chews, suction feeders, and parasites, suggest that some feeding methods were more influenced by seasonal changes than were others, e.g., parasites. Similarly, seasonality also appeared to impact the number of nematodes and the dominant genera employing many of the feeding methods. Changes in the abundance values of various feeding methods, and transitions from one dominant genus in a feeding method to another, both support the conclusion that season influenced the overall structure of the nematode community.

The most notable transitions in dominant genera within a feeding method included nematodes classified as deposit feeders, chewers, and suction feeders. During the transition from winter to spring, Monhystera replaced Terschellingia as the dominant deposit-feeding genus, and maintained this dominance throughout the remainder of the study. In addition, Semitobrilus was the dominant chewing nematode throughout both winter and spring; however, in summer, Iotonchus began to dominate throughout the remainder of the study. Dorylaimus was the dominant suction feeder in all seasons. This
trend suggests that an interaction between trophic levels may have been occurring in the system.

Both primary consumers (deposit and epistrate feeders) and secondary consumers (chewers and suction feeders) were observed. Over the course of the study, deposit feeder, chewer, and suction feeder abundance increased while epistrate feeders peaked in spring, then steadily declined. These patterns appear to be directly related to trophic interactions between the feeding methods. In particular, as prey item abundance increased, so too did predator abundance. Furthermore, this trend should hypothetically continue as long as the prey items remain abundant enough to support the predators (bottom-up regulation), or until the predators negatively impact the abundance of prey items (top-down regulation).

Both Small (1987) and Khan and Kim (2007) described *Iotonchus*, a chewer, and *Dorylaimus*, a suction feeder, as regularly consuming nematodes as primary sources of nutrients and energy. Furthermore, Shafquat et al. (1987) and Michiels and Traunspurger (2004) showed that as prey numbers increased, so too did the numbers of dorylaimid predators. Thus, it is reasonable to conclude that the increase in predators, i.e., chewers and suction feeders, was the result of an increase in the deposit and epistrate feeding prey items.

This explanation is sufficient for describing the trend observed during the first half of the study in winter and spring, but it does not fully explain the trends observed in summer and autumn concerning deposit feeders and epistrate feeders (Fig. V-5). In particular, deposit feeder presence continued to increase even in the presence of increasingly abundant predators, and only epistrate feeders appeared to be negatively
impacted by this same increase in predator numbers. Ostensibly, this interaction may simply appear to be bottom-up regulation, where prey items are so abundant in the system that the predators essentially render no negative impact on their numbers. Although this may indeed be the case, another explanation seems more appropriate for driving this pattern.

Bilgrami et al. (1986) and the review by Small (1987) both support the notion that mononchid predators, in which *Iotonychus* is included, are prey generalists. Furthermore, Yeates (1987) showed that predatory nematodes often feed on different prey at different stages of their development. This transition from one food item to another is tied to increased energetic demands incurred as the predators increase in size (Wardle et al. 1995). However, a similar size increase is not often observed in many prey items, such as deposit feeders who are restricted by the size of their food (bacteria and small unicellular eukaryotes) and, therefore, they remain relatively small throughout development. Thus, predators cannot be sustained by the same food items they consumed when they were smaller, and they must switch to a more substantial energy source, namely a larger nematode or other prey item. This appeared to occur here. As the study progressed, the predators in the ecosystem probably switched from consuming deposit feeders early, to epistrate feeders near the end. This switch consequently lead to a continued increase in abundance of the smaller deposit feeders, and a decrease in abundance of the larger epistrate feeders.

Additional studies aimed at understanding the influence of season on free-living nematode community composition should attempt to better understand the degree of interaction that these meiobenthos have with other fauna in the system at various times.
throughout the year. There appeared to be a clear interaction between trophic groups; however, due to the wide variety of food items that these organisms utilize (Bilgrami et al. 1986, Small 1987, Traunspurger 1997, Bilgrami and Gaugler 2004), as well as the important influence that they have on decomposition (Traunspurger et al. 1997) and primary production (Montagna 1995, Peters et al. 2007), it is naive to assume that nematodes alone are responsible for driving the patterns observed in this lake. A more in-depth look at the influence of these organisms on the organisms around them, as well as the impact of other organisms on the free-living nematodes described here, would help better explain the seasonal trends that were observed.

**Influence of substratum type**

Substratum type appeared to play a role in organizing the community structure of free-living nematodes in this system, such that two distinct nematode communities were observed. The first was observed in leaf and mixed substrata, and the second was observed in mud/algae and mud/clay substrata. In both cases, substratum type seemed to have the greatest influence on the prevalence and mean abundance of suction feeders and chewers. Deposit feeders, epistrate feeders, and parasitic nematodes showed no significant differences among substrata based on either prevalence or mean abundance. Chewers and suction feeders, on the other hand did show significant differences based on substratum type. Chewers were significantly more abundant in mixed substratum than any of the other substrata, and also showed a peak in prevalence in this substratum type. *Iotonchus* had the highest relative abundance of any other chewer in the mixed substratum type. Mixed substratum also exhibited the largest relative abundance of the deposit feeding *Monhystera*. It is unclear to what extent the presence of this potential
prey item might have influenced the spike in chewing nematode prevalence and
abundance. To further explore this question, the relative contribution of the major genera
contributing to this feeding method were considered in each substrata.

There were four major genera that contributed the majority of nematodes
demonstrating this feeding method. Of these, two were known predators (*Iotonchus* and
*Neotobrilus*) and the other two (*Semitobrilus* and *Prionchulus*) were described as either
predators or unicellular eukaryote feeders (Yeates et al. 1993). Interestingly, three of the
four dominated at least 1 of the substratum types. The relative abundance of *Iotonchus*
was highest in mixed and mud/algae substrata, *Neotobrilus* was highest in leaf substratum,
and, in mud/clay substratum, *Semitobrilus* had the highest relative abundance. The
overall variability in the dominant chewer genera from one substratum to the next, and
the relative stability of the deposit feeding nematodes (namely from the genus
*Mohnystera*) in all substrata, suggest that it is unlikely that a predator-prey interaction is
the driving force behind the site preference of chewing nematodes in the lake.

Suction feeders also appeared to be influenced by substratum type, though like
chewers, the actual cause of this influence is unclear. Nematodes demonstrating this
feeding type were significantly less abundant in leaf substratum than in all other substrata
and more abundant in mixed than in mud/clay substrata. Because the most abundant
suction-feeding genus was *Dorylaimus*, and this genus is known to be omnivorous
(Yeates et al. 1993), it seems reasonable that these individuals might prefer the most
diverse substratum type as it contains elements of leaf, mud/clay, and mud/algae substrata.
This sort of habitat could conceivably support a wider array of potential food items, both
floral and faunal.


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**Fig. V-1:** Illustration of buccal morphologies observed in the current study, as proposed by Traunspurger (1997). A and B: deposit feeders, C and D: chewers, E: epistrate feeder, F: suction feeder.
Table V-1: List of all genera observed based on the feeding method they employ. Values are expressed in the total number of individuals observed belonging to each genus (n), the total number of individuals presumed to employ each feeding method (total), and the relative abundance of each feeding method in the system (%).

<table>
<thead>
<tr>
<th>Feeding Method</th>
<th>n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Deposit Feeder/Swallow</td>
<td></td>
</tr>
<tr>
<td>Monhystera</td>
<td>683</td>
</tr>
<tr>
<td>Tarschellingia</td>
<td>60</td>
</tr>
<tr>
<td>Theiasomonhystera</td>
<td>23</td>
</tr>
<tr>
<td>Eumonhystera</td>
<td>22</td>
</tr>
<tr>
<td>Rhabdoclamus</td>
<td>17</td>
</tr>
<tr>
<td>Antiplectus</td>
<td>16</td>
</tr>
<tr>
<td>Panapriclamus</td>
<td>11</td>
</tr>
<tr>
<td>Chronogaster</td>
<td>9</td>
</tr>
<tr>
<td>Plectus</td>
<td>8</td>
</tr>
<tr>
<td>Paraplectonema</td>
<td>6</td>
</tr>
<tr>
<td>Monhystella</td>
<td>3</td>
</tr>
<tr>
<td>Alaimus</td>
<td>2</td>
</tr>
<tr>
<td>Rhabditoides</td>
<td>2</td>
</tr>
<tr>
<td>Paraphanomatamus</td>
<td>1</td>
</tr>
<tr>
<td>Cephalohus</td>
<td>1</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>854 (44.8%)</strong></td>
</tr>
<tr>
<td>Chewer</td>
<td></td>
</tr>
<tr>
<td>Itonchus</td>
<td>316</td>
</tr>
<tr>
<td>Semitobrilus</td>
<td>111</td>
</tr>
<tr>
<td>Neotobrilus</td>
<td>106</td>
</tr>
<tr>
<td>Prionchulus</td>
<td>86</td>
</tr>
<tr>
<td>Otonchus</td>
<td>10</td>
</tr>
<tr>
<td>Chrysonevroides</td>
<td>2</td>
</tr>
<tr>
<td>Paramononchus</td>
<td>1</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>632 (34.6%)</strong></td>
</tr>
<tr>
<td>Suction Feeder</td>
<td></td>
</tr>
<tr>
<td>Doryclamrus</td>
<td>273</td>
</tr>
<tr>
<td>Lelechus</td>
<td>26</td>
</tr>
<tr>
<td>Zeimura</td>
<td>1</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>300 (15.6%)</strong></td>
</tr>
<tr>
<td>Epistrate Feeder</td>
<td></td>
</tr>
<tr>
<td>Monochromadora</td>
<td>92</td>
</tr>
<tr>
<td>Prismonolamusa</td>
<td>19</td>
</tr>
<tr>
<td>Onccholamusa</td>
<td>8</td>
</tr>
<tr>
<td>Ironus</td>
<td>3</td>
</tr>
<tr>
<td>Trypia</td>
<td>2</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>124 (6.4%)</strong></td>
</tr>
<tr>
<td>Parasitic</td>
<td></td>
</tr>
<tr>
<td>Mermithida</td>
<td>8</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>8 (0.4%)</strong></td>
</tr>
</tbody>
</table>
Table V-2: Prevalence of various feeding methods for the overall system, for each season, and for each substratum type considered. Columns represent the percent of the total number of sites in each sample containing at least one individual employing said feeding method. DF/S=deposit feeder/swallower, C=chewer, SF=suction feeder, EF=epistrate feeder, and P=parasitic.

<table>
<thead>
<tr>
<th>Feeding Method</th>
<th>DF/S</th>
<th>C</th>
<th>SF</th>
<th>EF</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overall</td>
<td>48.7</td>
<td>30.9</td>
<td>31.2</td>
<td>11.7</td>
<td>1.9</td>
</tr>
<tr>
<td>Season</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Winter</td>
<td>36.4</td>
<td>18.2</td>
<td>9.1</td>
<td>9.1</td>
<td>0.0</td>
</tr>
<tr>
<td>Spring</td>
<td>38.1</td>
<td>25.0</td>
<td>16.7</td>
<td>13.1</td>
<td>0.0</td>
</tr>
<tr>
<td>Summer</td>
<td>48.8</td>
<td>27.4</td>
<td>29.8</td>
<td>12.5</td>
<td>1.2</td>
</tr>
<tr>
<td>Autumn</td>
<td>59.4</td>
<td>43.8</td>
<td>49.0</td>
<td>9.4</td>
<td>5.2</td>
</tr>
<tr>
<td>Substratum Type</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leaf</td>
<td>51.1</td>
<td>36.7</td>
<td>14.4</td>
<td>7.8</td>
<td>1.1</td>
</tr>
<tr>
<td>Mixed</td>
<td>55.1</td>
<td>42.3</td>
<td>38.5</td>
<td>11.5</td>
<td>5.1</td>
</tr>
<tr>
<td>Mud/Algae</td>
<td>42.9</td>
<td>26.5</td>
<td>39.8</td>
<td>15.3</td>
<td>1.0</td>
</tr>
<tr>
<td>Mud/clay</td>
<td>47.3</td>
<td>20.4</td>
<td>32.3</td>
<td>11.8</td>
<td>1.1</td>
</tr>
</tbody>
</table>
**Fig. V-2:** Prevalence of nematodes utilizing various feeding methods to obtain food items based on season.
**Fig. V-3:** Prevalence of nematodes utilizing various feeding methods to obtain food items based on substratum type.
Fig. V-4: Mean abundance based on feeding method. Error bars are standard error. Monte Carlo simulation analyses were used to determine whether feeding methods differed significantly from one another. P-values can be found in Appendix B-4.
Mean Nematode Abundance

Feeding Method

Chewer  Deposit Feeder/Swallower  Epistrate Feeder  Suction Feeder  Parasitic

Mean Nematode Abundance
**Fig. V-5:** Mean abundance of feeding methods based on season. Error bars are standard error. Monte Carlo simulation analyses were used to determine whether feeding methods differed significantly from one another within each season. P-values can be found in Appendix B-5.
Fig. V-6: Mean abundance of feeding methods based on substratum type. Error bars are standard error. Monte Carlo simulation analyses were used to determine whether feeding methods differed significantly from one another within each substratum type. P-values can be found in Appendix B-7.
CHAPTER VI

SUMMARY

Nematodes were among the most prevalent and abundant benthic invertebrates observed in Mallard Lake. Similar results have been reported in a number of systems including both lotic (Anderson 1992, Eisendle 2008) and lentic (Vidaković & Bogut 2004, Bergtold & Traunspurger 2005) habitats. Monhystera, classified as a bacterivore and deposit feeder/swallower in the current study, dominated nearly every month, season, and substratum type. This genus has been reported as the most dominant in a number of studies (Prejs 1977, Strayer 1985, Wu and Liang 1999, and Eisendle 2008). The family to which this genus belongs, Monhysteridae, has been reported to dominate a range of eutrophic ecosystems as well (Prejs 1977, Michiels and Traunspurger 2004, Bert et al. 2007). The great abundance of bacterivorous, deposit-feeding nematodes implies the important role they play in transferring energy into the food chain. These bacterivorous nematodes are then consumed by other benthic invertebrates, continuing the energy flow from primary producers to other organisms in the system. The seasonal changes in prevalence and abundance, especially in regards to predatory groups and their prey, supports the notion that that free-living nematodes carry out critical processes necessary for the success of the freshwater systems they inhabit.

Scale appeared to play a crucial role in determining which factors influenced the community structure of free-living nematodes in Mallard Lake. Temporal factors, specifically month and season, had the greatest influence on the community structure on a global scale, i.e., at the level of the lake. In contrast, substratum type appeared to have
little influence on the community structure at this scale, although mean abundance would suggest that, in general, the free-living nematodes preferred the mixed substratum when compared to the other substrata.

To achieve finer resolution of the community, all free-living nematodes collected were separated into 2 independent feeding classifications. The first focused on the type of food consumed by each genus observed in the system, while the second emphasized the method of food acquisition, which was based on buccal morphology. When the same system was viewed from these finer scales, it became clear that season, trophic level, and substratum all played important roles in structuring the free-living nematode community. Ultimately, it was concluded that the distribution and abundance of nematodes in Mallard Lake was driven by the influence of all 3 of these parameters. It remains unclear which factors most influence the system, but perhaps being able to answer this question is not nearly as important as the understanding that multiple factors are responsible, at least in part, for the patterns observed in the system.

These findings have implications for future, community-level analyses, and are applicable to any number of ecological studies interested in answering the same types of questions explored here. Perhaps the most important issue to consider in this type of study is scale (both spatial and temporal). These present results make clear that elucidating the role of various factors is directly related to the degree of inclusiveness, or exclusiveness, of the scale being used. Additionally, it is important to consider both spatial and temporal components in this type of study, since focusing on any single factor may lead to incorrect conclusions. The present study provides a good example of the complexity of field work, and makes clear the importance of a thorough understanding of
the factors (both biotic and abiotic) that may be influencing the system, and determining a scale (both spatial and temporal) from which to observe the system of interest.
LITERATURE CITED


APPENDICES

APPENDIX A (additional figures and tables):

A-1: Substratum temperatures over the course of the study (January to October, 2009). Dotted lines represent naturally occurring breaks in seasons.
**A-2:** Seasons broken down by the first and last collection date in each. Sample sizes in each season are also included.

<table>
<thead>
<tr>
<th>Season</th>
<th>First Collection</th>
<th>Last Collection</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>Summer</td>
<td>6/29/2009</td>
<td>8/18/2009</td>
<td>166</td>
</tr>
</tbody>
</table>
A-3: Mean seasonal abundance based on each feeding type observed. Error bars are standard error. Significance of differences between means within feeding types were determined by comparing standard error.
APPENDIX B (p-values for comparison of means):

**B-1:** Results of pairwise comparisons of mean abundance values based on mo for the entire data set (feeding type and method not considered separately). Corresponding p-values represent the averaged results of three Monte Carlo randomizations consisting of 10,000 iterations each (30,000 total iterations). The significance threshold was set at \( p \leq 0.05 \). Asterisks indicate that differences in means are significant.

<table>
<thead>
<tr>
<th>Month</th>
<th>January</th>
<th>February</th>
<th>March</th>
<th>April</th>
<th>May</th>
<th>June</th>
<th>July</th>
<th>August</th>
<th>September</th>
<th>October</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.233</td>
<td>0.310</td>
<td>0.184</td>
<td>0.264</td>
<td>0.267</td>
<td>0.361</td>
<td>0.356</td>
<td>0.000*</td>
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</tr>
<tr>
<td></td>
<td>0.333</td>
<td>0.979</td>
<td>0.512</td>
<td>0.234</td>
<td>0.356</td>
<td>0.006*</td>
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<td>0.000*</td>
<td>0.000*</td>
<td>0.000*</td>
</tr>
<tr>
<td></td>
<td>0.033*</td>
<td>0.025*</td>
<td>0.008*</td>
<td>0.000*</td>
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<td>0.039</td>
<td>0.039*</td>
<td>0.039*</td>
<td>0.039*</td>
</tr>
</tbody>
</table>

\[ \text{Month} \]
**B-2:** Results of pairwise comparisons of mean abundance values based on season for the entire data set (feeding type or method not considered separately). Corresponding p-values represent the averaged results of three Monte Carlo randomizations consisting of 10,000 iterations each (30,000 total iterations). The significance threshold was set at \( p \leq 0.05 \). Asterisks indicate that differences in means are significant.

<table>
<thead>
<tr>
<th>Season</th>
<th>Winter</th>
<th>Spring</th>
<th>Summer</th>
<th>Autumn</th>
</tr>
</thead>
<tbody>
<tr>
<td>Winter</td>
<td>--</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Spring</td>
<td>0.031*</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Summer</td>
<td>0.004*</td>
<td>0.442</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>Autumn</td>
<td>0.000*</td>
<td>0.049*</td>
<td>0.047*</td>
<td>--</td>
</tr>
</tbody>
</table>
**B-3:** Results of pairwise comparisons of means based on substratum type for the entire data set (feeding type and method not considered separately). Corresponding p-values represent the averaged results of three Monte Carlo randomizations consisting of 10,000 iterations each (30,000 total iterations). The significance threshold was set at $p \leq 0.05$. Asterisks indicate that differences in means are significant.

<table>
<thead>
<tr>
<th>Substratum Type</th>
<th>Leaf</th>
<th>Mixed</th>
<th>Mud/Algae</th>
<th>Mud/Clay</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leaf</td>
<td>--</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mixed</td>
<td>0.000*</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mud/Algae</td>
<td>0.388</td>
<td>0.000*</td>
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<td></td>
</tr>
<tr>
<td>Mud/Clay</td>
<td>0.361</td>
<td>0.000*</td>
<td>0.239</td>
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</tr>
</tbody>
</table>
**B-4**: Results of pairwise comparisons of means for nematodes utilizing different feeding methods for primary nutrient acquisition. Corresponding p-values represent the averaged results of three Monte Carlo randomizations consisting of 10,000 iterations each (30,000 total iterations). The significance threshold was set at $p \leq 0.05$. Asterisks indicate that differences in means are significant. DF/S=deposit feeder/swallow, C=chewer, SF=suction feeder, EF=epistrate feeder, and P=parasitic.

<table>
<thead>
<tr>
<th>Feeding Method</th>
<th>DF/S</th>
<th>C</th>
<th>SF</th>
<th>EF</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>DF/S</td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>0.026*</td>
<td>--</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SF</td>
<td>0.000*</td>
<td>0.002*</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>EF</td>
<td>0.000*</td>
<td>0.000*</td>
<td>0.034*</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>P</td>
<td>0.000*</td>
<td>0.000*</td>
<td>0.004*</td>
<td>0.231</td>
<td>--</td>
</tr>
</tbody>
</table>
**B-5:** Results of pairwise comparisons of means between nematodes classified by feeding methods broken down by season. Corresponding p-values represent the averaged results of three Monte Carlo randomizations consisting of 10,000 iterations each (30,000 total iterations). The significance threshold was set at \( p \leq 0.05 \). Asterisks indicate that differences in means are significant. DF/S=deposit feeder/swallower, C=chewer, SF=suction feeder, EF=epistrate feeder, and P=parasitic.

<table>
<thead>
<tr>
<th>Season</th>
<th>Feeding Method</th>
<th>C</th>
<th>SF</th>
<th>EF</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>SPRING</td>
<td>DFS</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td></td>
<td>0.000*</td>
<td>0.004*</td>
<td></td>
<td></td>
</tr>
<tr>
<td>AUTUMN</td>
<td>DFS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.000*</td>
<td>0.000*</td>
<td></td>
<td>0.015*</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.000*</td>
<td>0.000*</td>
<td></td>
<td>0.017*</td>
</tr>
<tr>
<td>WINTER</td>
<td>DFS</td>
<td></td>
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</tr>
<tr>
<td></td>
<td></td>
<td>0.020*</td>
<td>0.337</td>
<td>0.348</td>
<td>0.419</td>
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<tr>
<td></td>
<td></td>
<td>0.037*</td>
<td>0.473</td>
<td>0.333</td>
<td></td>
</tr>
<tr>
<td>SUMMER</td>
<td>DFS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.200</td>
<td>0.081</td>
<td>0.127</td>
<td>0.563</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.008*</td>
<td>0.002*</td>
<td></td>
<td>0.072</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.000*</td>
<td>0.000*</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Results of pairwise comparisons of means between seasons broken down by
nematodes classified by feeding method. Corresponding p-values represent the averaged
results of three Monte Carlo randomizations consisting of 10,000 iterations each (30,000
total iterations). The significance threshold was set at $p \leq 0.05$. Asterisks indicate that
differences in means are significant.

<table>
<thead>
<tr>
<th></th>
<th>CHEWER</th>
<th></th>
<th>DEPOSIT FEEDER/SWALLOWER</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Winter</td>
<td>Spring</td>
<td>Summer</td>
<td>Autumn</td>
</tr>
<tr>
<td>Season</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Winter</td>
<td>--</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Spring</td>
<td>0.334</td>
<td>--</td>
<td>0.413</td>
<td>--</td>
</tr>
<tr>
<td>Summer</td>
<td>0.376</td>
<td>0.413</td>
<td>--</td>
<td>0.404*</td>
</tr>
<tr>
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<td>0.084</td>
<td>0.084</td>
<td>0.040*</td>
<td>--</td>
</tr>
</tbody>
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<table>
<thead>
<tr>
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<th></th>
<th>SUCTION FEEDER</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Winter</td>
<td>Spring</td>
<td>Summer</td>
<td>Autumn</td>
</tr>
<tr>
<td>Season</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Winter</td>
<td>--</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Spring</td>
<td>0.117</td>
<td>--</td>
<td>0.104</td>
<td>--</td>
</tr>
<tr>
<td>Summer</td>
<td>0.666</td>
<td>0.104</td>
<td>--</td>
<td>0.490</td>
</tr>
<tr>
<td>Autumn</td>
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<td>0.047*</td>
<td>0.490</td>
<td>--</td>
</tr>
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<table>
<thead>
<tr>
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<th></th>
</tr>
</thead>
<tbody>
<tr>
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<td>Spring</td>
</tr>
<tr>
<td>Season</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Winter</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Spring</td>
<td>0.329</td>
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</tr>
<tr>
<td>Summer</td>
<td>0.736</td>
<td>0.374</td>
</tr>
<tr>
<td>Autumn</td>
<td>0.011*</td>
<td>0.006*</td>
</tr>
</tbody>
</table>
**B-7:** Results of pairwise comparisons of means between nematodes classified by feeding methods broken down by substratum type. Corresponding p-values represent the averaged results of three Monte Carlo randomizations consisting of 10,000 iterations each (30,000 total iterations). The significance threshold was set at $p \leq 0.05$. Asterisks indicate that differences in means are significant. DF/S=deposit feeder/swallower, C=chewer, SF=suction feeder, EF=epistrate feeder, and P=parasitic.

<table>
<thead>
<tr>
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<th>MUD/CLAY</th>
<th>MUD/ALGAE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DFS</td>
<td>C</td>
<td>SF</td>
<td>EF</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.151</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.006</td>
<td>0.005*</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
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<tr>
<td></td>
<td></td>
<td>0.000</td>
<td>0.000*</td>
<td>0.000*</td>
</tr>
</tbody>
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<table>
<thead>
<tr>
<th>Feeding Method</th>
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<th>MUD/ALGAE</th>
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</thead>
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<td>C</td>
</tr>
<tr>
<td></td>
<td>0.004*</td>
<td>0.000*</td>
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<tr>
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<td>0.000*</td>
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</tr>
<tr>
<td></td>
<td>0.000*</td>
<td>0.398</td>
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</table>

<table>
<thead>
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<th>Feeding Method</th>
<th>LEAF</th>
<th>MUD/ALGAE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DFS</td>
<td>C</td>
</tr>
<tr>
<td></td>
<td>0.010*</td>
<td>0.000*</td>
</tr>
<tr>
<td></td>
<td>0.000*</td>
<td>0.031*</td>
</tr>
<tr>
<td></td>
<td>0.000*</td>
<td>0.145*</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Feeding Method</th>
<th>LEAF</th>
<th>MUD/ALGAE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DFS</td>
<td>C</td>
</tr>
<tr>
<td></td>
<td>0.010*</td>
<td>0.000*</td>
</tr>
<tr>
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<td>0.000*</td>
<td>0.031*</td>
</tr>
<tr>
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<td>0.000*</td>
<td>0.145*</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Feeding Method</th>
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<th>MUD/ALGAE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DFS</td>
<td>C</td>
</tr>
<tr>
<td></td>
<td>0.010*</td>
<td>0.000*</td>
</tr>
<tr>
<td></td>
<td>0.000*</td>
<td>0.031*</td>
</tr>
<tr>
<td></td>
<td>0.000*</td>
<td>0.145*</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Feeding Method</th>
<th>LEAF</th>
<th>MUD/ALGAE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DFS</td>
<td>C</td>
</tr>
<tr>
<td></td>
<td>0.010*</td>
<td>0.000*</td>
</tr>
<tr>
<td></td>
<td>0.000*</td>
<td>0.031*</td>
</tr>
<tr>
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<td>0.000*</td>
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<table>
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<th>Feeding Method</th>
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<th>MUD/ALGAE</th>
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</thead>
<tbody>
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</tr>
<tr>
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<td>0.000*</td>
</tr>
<tr>
<td></td>
<td>0.000</td>
<td>0.031*</td>
</tr>
<tr>
<td></td>
<td>0.000</td>
<td>0.145*</td>
</tr>
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</table>
### Results of pairwise comparisons of means between substrata broken down by nematodes classified by feeding method. Corresponding p-values represent the averaged results of three Monte Carlo randomizations consisting of 10,000 iterations each (30,000 total iterations). The significance threshold was set at $p \leq 0.05$. Asterisks indicate that differences in means are significant.

<table>
<thead>
<tr>
<th>CHEWER Substratum Type</th>
<th>Leaf</th>
<th>Mixed</th>
<th>Mud/Algae</th>
<th>Mud/Clay</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leaf</td>
<td>--</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mixed</td>
<td>0.003*</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mud/Algae</td>
<td>0.475</td>
<td>0.002*</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>Mud/Clay</td>
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<td>0.333</td>
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</tr>
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<table>
<thead>
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<th>Mud/Algae</th>
<th>Mud/Clay</th>
</tr>
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<tbody>
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<td>Leaf</td>
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</tr>
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<td>Mud/Algae</td>
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<tr>
<td>Mud/Clay</td>
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<td>0.363</td>
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<table>
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<th>Mud/Clay</th>
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<td></td>
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<tr>
<td>Mud/Clay</td>
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<th>Mud/Algae</th>
<th>Mud/Clay</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leaf</td>
<td>--</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mixed</td>
<td>0.000*</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mud/Algae</td>
<td>0.002*</td>
<td>0.128</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>Mud/Clay</td>
<td>0.009*</td>
<td>0.350*</td>
<td>0.259</td>
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<table>
<thead>
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<th>PARASITIC Substratum Type</th>
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<th>Mud/Algae</th>
<th>Mud/Clay</th>
</tr>
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<tbody>
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<td>Leaf</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mixed</td>
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<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mud/Algae</td>
<td>0.376</td>
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<td></td>
</tr>
<tr>
<td>Mud/Clay</td>
<td>0.386</td>
<td>0.072</td>
<td>0.565</td>
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</table>
APPENDIX C:

C-1: List of genera identified in the current study. Taxonomy based on De Ley et al. (2006) unless otherwise specified.

<table>
<thead>
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<th>Family</th>
<th>Genus</th>
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<td>Rhabdolaimus*</td>
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<tr>
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<td>Qudsanematida</td>
<td>Chrysonemoides</td>
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<tr>
<td></td>
<td>Dorylaimida</td>
<td>Dorylaimus</td>
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<td>Alaimida</td>
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<td></td>
<td>Ironidae</td>
<td>Ironus</td>
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<td>Oncholaimida</td>
<td>Oncholaimus</td>
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<td>Merrittida</td>
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<td>Mononchida</td>
<td>Oionchus</td>
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<td>Aphanolaimida</td>
<td>Paraphanolaimus</td>
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* based on taxonomy proposed by Yeates et al. (1993)
† based on description by Timm (1964)

P can be parasitic, mostly in plants or insects

Taxonomy based on De Ley et al (2006)
**APPENDIX D** (species accumulation curves):

**D-1:** Species accumulation curves for mo based on the number of sites sampled. Curves were generated using the outputs generated by the $S_{obs}$ (Mao Tau) function (for determining the expected species richness) in the EstimateS software package.
Number of Sites Sampled vs. Number of Species

- March
- April
- May
- June
- July
- August
- September
- October

March: 0 to 35
April: 0 to 35
May: 0 to 35
June: 0 to 35
July: 0 to 35
August: 0 to 35
September: 0 to 35
October: 0 to 35

Number of Sites Sampled: 1 to 65
**D-2:** Species accumulation curves for seasons based on the number of sites sampled. Curves were generated using the outputs generated by the $S_{obs}$ (Mao Tau) function (for determining the expected species richness) in the EstimateS software package.
D-3: Species accumulation curves for substratum type based on the number of sites sampled. Curves were generated using the outputs generated by the $S_{obs}$ (Mao Tau) function (for determining the expected species richness) in the EstimateS software package.
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