Responses of *Diaptomus* spp. from an oligotrophic lake to variations in food quality

By

Nancy M. Butler

B.A., The University of Colorado, 1979
M.A., The University of Colorado, 1983

A THESIS SUBMITTED IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR THE DEGREE OF
DOCTOR OF PHILOSPHOPHY

in

THE FACULTY OF GRADUATE STUDIES

(Department of Zoology)

We accept this thesis as conforming to
the required standard

THE UNIVERSITY OF BRITISH COLUMBIA

October 1990

In presenting this thesis in partial fulfilment of the requirements for an advanced degree at the University of British Columbia, I agree that the Library shall make it freely available for reference and study. I further agree that permission for extensive copying of this thesis for scholarly purposes may be granted by the head of my department or by his or her representatives. It is understood that copying or publication of this thesis for financial gain shall not be allowed without my written permission.

Department of Zoology

The University of British Columbia
Vancouver, Canada

Date 12 October 1990

ABSTRACT

Copepods live in a nutritionally dilute environment, experiencing temporal and spatial variations in food supply which differ in magnitude and predictability. Understanding the mechanisms by which organisms deal with changes in their food is a primary concern in elucidating the nutritional ecology of zooplankton and the role of food in structuring zooplankton communities. In this thesis, I examine changes in behavior, morphology, and physiology of two species of calanoid copepods (*Diaptomus kenai* and *D. leptopus*) in response to variation in food composition and density.

In Chapter Two, I present a study of population-level responses to variation in food composition and quality, using fertilization techniques to generate a range of phytoplankton communities in field enclosures. The phytoplankton assemblages studied supported copepod populations which differed in such attributes as population size, reproduction, and body size. The most striking finding of this study was the occurrence of two co-existing size classes of *D. kenai*, the abundance and clutch size of which varied among the enclosures, suggesting differences between the two classes in their ability to utilize the different phytoplankton communities. Chapter Three investigates patterns of lipid storage in response to changes in food supply. I concluded that lipid stores were affected by species composition of the phytoplankton food and the two copepod species differed in their sensitivity to differences in cell chemistry. Chapter Four investigates behavioral responses of the two size classes of *D. kenai* to changes in food composition and abundance. Subtle differences in feeding behavior suggest that the two sizes differ in their utilization of available food.

These results demonstrate that *D. kenai* and *D. leptopus* are capable of responding to changes in their food supply through modifications of their behavior, morphology, and physiology over a range of magnitudes and time scales. There can be very subtle changes in feeding behavior or very pronounced changes in size structure. Responses occur over time scales ranging

from hours to days to seasons. These results also bring into question the utility of models generated under laboratory conditions in predicting behaviors or dynamics of copepod populations and communities in nature.

Table of Contents

List of Tables

List of Figures

ACKNOWLEDGEMENTS

This thesis would never have come to see the light of day without the support of Bill Neill. As my supervisor, Bill was generous in providing encouragement, harassment, and money to keep both me and the research going. He was always there when I needed him and left me to my own errors when I thought I didn't need him. Thus, I take full responsibility for the shortcomings of this thesis but am indebted to Bill for its strengths. I also thank Carl Walters, Paul J. Harrison, Al Lewis, and Tom Carefoot who, as members of my supervisory committee, provided both intellectual stimulation and moral support throughout the course of my research. My research was supported by a Natural Science and Engineering Research Council of Canada Operating Grant to William E. Neill and by a University of British Columbia Graduate Fellowship.

Use of the Iatroscan was made possible through the generosity of Dr. Wayne Gardner at the Great Lakes Environmental Research Laboratory in Ann Arbor, Michigan. Wayne also provided expert guidance as I learned the lipid extraction procedure. During his stay at GLERL, Dr. Chris Parrish unselfishly parted with his time and knowledge to assist me in the use of the Iatroscan.

Numerous friends and colleagues at UBC and the now defunct Institute of Animal Resource Ecology provided intellectual support and comic relief throughout the course of my stay at UBC; I would particularly like to acknowledge Dan Brooks, Danuta Dolecki, Jon Harrison, Jill Lancaster, Debbie McLennan, Daniel Schindler, Curtis Suttle, and the highly acclaimed (and extremely obscure) Ralph Yorque.

Finally, I am forever indebted to Jim Lowden and the *Feng Shui* for helping to keep things in perspective, to Posy and Poppy, without whose help this thesis would have been done in half the time, and to Carl Walters for picking up the pieces and putting them back together again.

And NUH is the letter I use to spell Nutches
Who live in small caves, known as Nitches, for hutches.
These Nutches have troubles, the biggest of which is
The fact there are many more Nutches than Nitches.
Each Nutch in a Nitch knows that some other Nutch
Would like to move into his Nitch very much.
So each Nutch in a Nitch has to watch that small Nitch
Or Nutches who haven't got Nitches will snitch.

Dr. Seuss
"On Beyond Zebra"

Chapter One

INTRODUCTION

Temperate lakes are variable environments characterized by fluctuations over a wide range of physical, chemical, and biological parameters. Produced by external (e.g., weather, erosion, and human activities) and internal dynamics (e.g., circulation patterns, food web interactions), this variation both creates opportunities for, and sets constraints upon, the biological components. Through natural selection, the physiology, behavior, morphology, and life history of organisms are modified to cope with the variation characteristic of their habitat. Planktonic invertebrate herbivores must deal with substantial variations in physical factors, food, and predation that fluctuate on time scales ranging from minutes to seasons or years, and spatial scales ranging from micrometers to meters. The characteristics of an organism will affect its ability to perceive and respond to such fluctuations. For example, as a consequence of differences in such attributes as feeding mechanisms, food preferences, and acclimation, zooplankton will differ in their perception of temporal and spatial variation in food quality and quantity (Donaghay 1988). This thesis concerns patterns of adjustment on different time scales among calanoid copepods to variations in the phytoplankton foods they need to survive, grow, and reproduce; it focuses on feeding, energy storage, and demographics of two coexisting species of calanoid copepod, *Diaptomus kenai* and *D. leptopus*.

The composition and productivity of the zooplankton community are dominated by three major taxonomic groups: rotifers, cladocerans, and copepods, all of which have both herbivorous and carnivorous representatives. The rotifers are primarily suspension feeders, ingesting food particles through the action of the coronal cilia, generally ingesting particles less than 12 μm (see review by Pourriot 1977), although some species, for example large predatory *Asplanchna*, can even ingest small crustaceans. In general, although individual species have a large food-size range, coexisting rotifer species can be separated along a food

size gradient (Makarewicz and Likens 1975), presumably minimizing competitive interactions. There are two cladoceran groups with predaceous members that feed by grasping their prey, but the majority of the cladocerans are suspension feeders. As a consequence of their feeding mechanism, ingestion is strongly correlated with food size (Brendelberger 1975; Lampert 1987), although there are suggestions that a degree of selectivity may occur (e.g., Butler *et al.* 1989).

Freshwater copepods are represented by three orders. One of these, the harpacticoids, are primarily benthic herbivores. The cyclopoid copepods, while containing some herbivorous representatives, are generally recognized as active predators, although there seems to be some disagreement as to whether they are classified as primarily limnetic (Pennak 1978) or littoral (Zaret 1980). Predatory cyclopoid copepods are characterized by an ontogenetic shift in feeding habits, being primarily herbivorous as nauplii and carnivorous as adults. The calanoid copepods have long been considered to be primarily herbivorous, non-selective filter feeders, although Pennak (1978) did allow that there was perhaps some evidence of omnivory. Calanoid copepods are now recognized as being not only omnivorous, but also highly effective predators (Anderson 1967, 1970; Williamson and Butler 1986; Williamson 1987; Williamson and Vanderploeg 1988) as well as highly discriminating feeders (e.g., Fulton 1988; Butler *et al.* 1989; DeMott 1989; Verity and Smayda 1989). Because a substantial portion of the zooplankton community is often composed of calanoid copepods, calanoid grazing and subsequent production may represent a substantial contribution to energy flow from primary producers (the phytoplankton) to higher trophic levels (Herzig *et al.* 1980; Evans 1986; Crowder *et al.* 1987; Black and Hairston 1988). Consequently, variation in their utilization of the phytoplankton can potentially have severe repercussions throughout the food web.

Changes in the phytoplankton community can be induced by a number of factors. Grazing by copepods can impact both size structure and species composition of the phytoplankton

community (Carpenter *et al.* 1987; Vanni 1987). Variations in composition and concentration of inorganic nutrients can also affect phytoplankton community structure. The link between nutrient levels and such phytoplankton community parameters as species composition, biomass, and diversity has been demonstrated in theoretical, lab, and field studies (see review by Kilham and Kilham 1984). The strong relationship between lake phosphorus concentration and phytoplankton production (Vollenweider 1976) suggests that many lakes are nutrient-limited, a notion further supported by demonstrated increases in phytoplankton production induced by nutrient enrichment (e.g., Schindler *et al.* 1973; Schindler and Fee 1974; Lane and Goldman 1984; Stockner and Shortreed 1985). Not only is total phytoplankton biomass affected by variation in nutrient levels, but also phytoplankton species composition. Lab studies have demonstrated that nutrient supply ratios may have a strong influence on the structure of phytoplankton communities (Tilman 1981; Kilham 1986; Turpin 1986; Tilman *et al.* 1986). These lab studies have been supported by field studies of Schindler (1977) and Barica *et al.* (1980), who induced variation in lake phytoplankton composition by altering the nitrogen:phosphorus ratios. Not only is the overall abundance of the phytoplankton related to nutrient levels, but also community diversity, which decreases as nutrient loading increases (Jones *et al.* 1978).

Natural sources of variation in nutrient supplies range from small-scale events lasting minutes (e.g., nutrient excretion by zooplankton: Lehman and Scavia 1982 a,b; Scavia *et al.* 1984) to large-scale seasonal events such as lake turnover (Wetzel 1983). Such variations may provide temporally and spatially separate niches, permitting the persistence of diverse assemblages of competing algal species (e.g., Richerson *et al.* 1970; Dickman and Efford 1972; Petersen 1975). For example, as spring turnover replenishes nutrients, there is often a bloom of small, highly edible algae, which are slowly replaced by a summer phytoplankton community dominated by less edible species (Reynolds 1984; Sommer *et al.* 1986). The frequency and extent of fluctuations can vary among environments. Eutrophic systems,

which tend to have high primary productivity, can undergo pronounced seasonal variation in phytoplankton community structure. Such changes can be attributed to seasonal variation in nutrient levels (Knoechel and Kalff 1978; Tilman 1982), grazing by zooplankton (Crumpton and Wetzel 1982; Sprules and Knoechel 1984), or differential nutrient cycling by the zooplankton (Lehman 1984). Oligotrophic systems, on the other hand, are characterized by low primary productivity and variation in the phytoplankton tends to occur on smaller temporal and spatial scales (e.g., Venrick 1990).

Thus, the phytoplankton community presents to zooplankton a variable food environment that fluctuates over a range of temporal and spatial scales. The potential consequences of a fluctuating environment can be conceptualized at three different levels (Schoener 1989): the individual level, reflected in behavioral, physiological, and morphological characteristics; the population level, manifested in population dynamics and equilibria; and, ultimately, the community level, reflecting interactions of the populations. With changes in food resources there can be changes in competitive ability (Jacobs 1977a,b; Vanni 1986, 1987; Hoenicke and Goldman 1987) or changes in feeding rate and diel migration patterns (Roman *et al.* 1988). Zooplankton may deal with spatial variation by migrating to food rich patches (Meyers 1984; Dagg 1985; Huntley 1985; Johnsen and Jakobsen 1987) or with temporal variation by utilizing energy stores (Paffenhöfer 1984).

Behavioral responses to variation in diet have long been popular theoretical and empirical research topics as researchers have become increasingly aware of the consistent relationship between food density and forager distribution (e.g., Jespersen 1924) and density (e.g., Taber 1956). We now recognize that the rate at which a forager obtains its food can vary in complex ways with food density, distribution, and composition. Early theoretical and experimental work by Solomon (1949), Holling (1959), and Ivlev (1961) described the "functional response" of a predator's feeding rate at various densities of a single prey type, with prey capture rate shown to increase with food density. The preference for any given

food is a consequence of the interactions of consumer and food characteristics affecting the processes of food detection, capture, and ingestion. Ivlev (1961) introduced the concept of selective feeding, or electivity, to quantify an organism's preference for the variety of food items available to it. By comparing the relative amount of a food type in the diet with its abundance in the environment, he generated an index by which the food items in a consumer's diet could be ranked. His original index has been modified by a number of researchers (incorporating a variety of factors, including number of alternative food choices, and relative and absolute differences in ingestion and availability) and the more commonly used of these have been reviewed by Lechowicz (1982).

Murdoch (1969) expanded the concept of selective feeding with predictions that a forager might "switch" preference for alternate food items as their relative abundance changes, similar to Tinbergen's (1960) theory of search-image mediated feeding, which suggests that search images originate from and are maintained by frequent encounters with the most abundant food. Contrary to predictions, however, Murdoch observed that when preference for a particular food item was strong, the forager continued to consume that item by an amount proportional to its abundance in nature (i.e. percent ingested did not change with food density). Further studies (Murdoch *et al.* 1975) suggested that switching between food items occurred only when food preference is weak. Landry (1981) observed that feeding in marine copepods occurred in a manner consistent with Murdoch's original (1969) predictions, but on a much larger scale. After a bloom of a single phytoplankton species, feeding rates on all phytoplankton species increased; as a result, feeding rates on rare species increased as the abundance of alternate species greatly increased. These results were contrary to the predictions of Murdoch (1969) but similar to the predictions of the foraging model of Engen and Stenseth (1984), which predicted that, when multiple choices are presented, food selection would be independent of ranking and of the availability of the different food items. However, Landry observed that, as the abundant species (and total phytoplankton) declined in density at the end of the bloom, rather than broadening diet range

(as MacArthur and Pianka 1966 would predict) or switching to concentrate on different phytoplankton species (as Murdoch 1969 would predict), the copepods switched foraging emphasis to a whole new trophic level: the zooplankton.

As with Landry's observations of copepod feeding behavior, observed foraging responses frequently differ from the predictions of foraging models, leading to criticisms that the utility of such models is restricted to situations similar to those from which the models were derived (e.g., Ranta and Nuutinen 1985). Rather than dismissing the models as ineffective due to their limited value as accurate predictors of foraging, it is far more interesting to consider possible reasons for deviations from the predictions. Krebs *et al.* (1983) and Abrams (1990) suggest that discrepancies between model predictions and observed responses may be attributed to adaptive foraging behavior differentially affecting the parameters of the model; that is, foragers can be adaptive and flexible, altering feeding behaviors in response to changes in resource abundance and composition. Thus, it is predicted that *Diaptomus* feeding behavior will vary as a function of temporal or even spatial variation in food.

Theories concerning selectivity and diet were expanded to consider food not only in terms of relative abundance but also in terms of food value. MacArthur and Pianka (1966), in their model predicting diet breadth, expressed food value as "profitability"; that is, the energy gain as a function of the energy spent locating, capturing, and ingesting a food item balanced by the energy value realized after ingestion. While physical characteristics of a food item such as cell size, cell shape, or overall abundance may affect the first three attributes, it is the internal, or chemical, composition of the cell that will determine its nutritional quality. The importance of nutritional content of food items has been demonstrated in a number of studies comparing such life history attributes as growth rate, survival, and reproduction in different food regimes. When larval mayflies were raised on five different foods over a range of temperatures, the size and weight of the resulting adults were influenced more by larval diet

than by development temperature (Sweeney *et al.* 1986). Similar results were reported for prawns (Jones *et al.* 1979), isopods (Carefoot 1984), oysters (Laing and Millican 1986), insects (Hansen *et al* 1983; Kondratieff and Simmons 1984; Cargill *et al* 1985), and freshwater (Warren *et al.* 1986) and marine (Huntley *et al.* 1987) copepods.

Nutritional value of a food item may also affect the internal chemistry of the forager. Variation in lipid content has been attributed to variations in food quality (i.e. chemical composition) for prawns (Whyte *et al.* 1986), oysters (Chu and Webb 1984; Gallager and Mann 1986), copepods (Håkanson 1987; Hagen 1988), amphipods (Clarke *et al.* 1985), fish (Benson *et al.* 1972), and rodents (Schemmel 1976). Lipid storage may also represent the consequences of interactions between energy input and utilization in a varying environment (Pianka 1976). Reviews by Lee *et al.* (1972), Benson *et al.* (1972), and Lawrence (1976) all describe variation in lipid stores in marine fishes and invertebrates as a function of food supply and energy demands.

Lipid energy stores have been documented for two major zooplankton groups, the marine copepods and the freshwater cladocerans (see the reviews of Benson *et al.* 1972; Lee *et al.* 1972; Lawrence 1976; Goulden and Henry 1984). There is also evidence of glycogen energy stores in non-feeding male rotifers (Wurdak and Gilbert 1980). Lipid stores serve two major functions. First, they are the sole energy source for non-feeding copepod nauplii (Lee *et al.* 1974; Benson and Lee 1975) and they are a supplemental energy source for juvenile cladocerans (Goulden and Henry 1984). Second, lipid stores may provide energy during periods of low food availability, either on a diurnal scale, such as encountered during vertical migration (Lee and Barnes 1975) or on a seasonal scale (Goulden and Henry 1984; Paffenhöfer 1984).

Lipids are generally visible in the body cavity as small scattered droplets or, as in the case of some marine copepods, as a single large sac. Lipids are stored either as triacylglycerides

(glycerol esters with three fatty acids) or as wax ester (an ester of fatty acids and fatty alcohol) (Hadley 1985). Arctic and deep sea copepods, which encounter short periods of food-rich conditions followed by long periods of low food abundance, primarily store wax esters, while triacylglycerides predominate in the lipids of surface dwelling copepods in temperate and tropical seas. Triacylglycerides are the major storage lipid for most freshwater copepods (Arts and Sprules 1987; Cavaletto *et al.* 1989) although some species store wax esters, apparently as a consequence of their relatively recent marine origin (Cavaletto *et al.* 1989).

The fatty acids making up triacylglycerides are most likely derived from the diet, as they cannot be directly synthesized (Conklin and Provasoli 1977; Henderson and Sargent 1980). Thus, the ability to accumulate lipids may be a function of diet composition as well as overall abundance. There is not only variation in fatty acid composition of algae according to the nutrient conditions under which the algae grow, but there is also interspecific variation in fatty acid composition. For example, cyanobacteria synthesize very few fatty acids, which may, in part, account for their poor nutritional quality as cladoceran food (Arnold 1971; Porter and Orcutt 1980). This notion is supported by the work of D'Abramo (1979), who reports that cladocerans maintained on artificial diets similar to cryptophytes or diatoms in fatty acid composition are significantly more productive than those maintained on a diet resembling the cyanobacteria in fatty acid composition.

The Objectives

The primary aim of this thesis is to expand our understanding of food web dynamics in nature by investigating and identifying the impacts of natural and induced variation in food supply upon physiological, behavioral, morphological, and life history characteristics. Calanoid copepods are excellent model organisms for this study not only for physiological reasons, given their sensitive discriminatory feeding abilities and with their ability to store lipids, but also for ecological reasons, as evidenced by their importance in aquatic

ecosystems.

In Chapter Two, I present the results of a field enclosure study investigating the effects of food composition on *Diaptomus* populations. The enclosures were fertilized over a range of nitrogen:phosphorus ratios to produce a variety of phytoplankton communities, and copepod populations were monitored for changes in such attributes as population density, body size, and clutch size among the different food regimes.

The second goal of the thesis is to identify some of the mechanisms by which variations in phytoplankton community structure affect the *Diaptomus* populations. In Chapter Three, I investigate the effects of inter- and intraspecific variations in food on the amount and composition of lipid stores. Copepods were removed from the enclosures studied in Chapter Two to monitor variation in lipid content among different phytoplankton communities. To investigate the effects of intraspecific variation in food quality, copepods were maintained on single species diets of either nitrogen-rich or nitrogen-limited algae.

Finally, Chapter 4 concentrates on the behavioral attributes of feeding as they relate to food composition and density to elucidate the nature of intraspecific variation in the responses of *Diaptomus kenai* to diet manipulations, as indicated in the results of Chapters Two and Three. Feeding rates and selectivities of *Diaptomus* were calculated for and compared among a variety of food regimes differing in species composition and density.

Chapter Two

DIAPTOMUS POPULATION DYNAMICS

Seasonal variation in egg production by copepods (Kimmerer 1984; Peterson 1985; Smith and Lane 1985; Crawford and Daborn 1986; Williamson and Butler 1987) and cladocerans (Hall 1964; Kerfoot 1974; Lampert 1988) suggests that reproduction is strongly influenced by variations in environmental conditions, particularly food. In addition, offspring survival can be strongly affected by variations in food concentration (e.g. Williamson *et al.* 1985). Because the critical factors controlling copepod populations are those affecting offspring survival and adult reproduction (Gehrs and Robertson 1975), effects of variation in food should be readily expressed in terms of population density when predation is not a controlling factor. This study reports on the effects of phytoplankton community structure on life history parameters of two species of calanoid copepod, *Diaptomus kenai* and *D. leptopus*, including changes in population density as well as changes in adult fecundity and body size.

With only a few exceptions, all species of copepod reproduce sexually, producing one (univoltine) or several (multivoltine) generations per year. After mating (see Blades-Eckelbarger 1986 for details), diaptomid females produce membrane-enclosed clutches of subitaneous eggs (those which hatch shortly after extrusion) or resting eggs (which hatch after a dormant period). In addition to species differences, fecundity can be a sensitive indicator of environmental conditions, varying with such factors as food availability, food quality, mate availability, and temperature (e.g., Marshall and Orr 1952; Paffenhöfer and Knowles 1978; Johnson 1980; Cahoon 1981).

In the course of developing from egg to adult, the copepod passes through six naupliar stages and six copepodite stages (the last being the adult), each accompanied by a

progressive increase in size with the addition of a body segment and a pair of appendages (see Björnberg 1986 for details). Copepods are sexually dimorphic at the adult stage. The genetic mechanism for sex determination remains unclear, but it is presumed that proportions of males and females within clutches start out equal, as evidenced by adult sex ratios of many species (see Davies 1984). However, development rate, survival, and spatial distribution may differ between males and females in some species (e.g., Marshall and Orr 1955; Corkett and McLaren 1978; Davies 1984) making it difficult to extrapolate clutch sex ratios from sex ratios of adult populations.

While laboratory studies of feeding can demonstrate the effects of very simple food regimes, the relationship between food and life history strategies in nature can be quite complex, with interactions between food quality and quantity obscuring diet effects (e.g., Abou Debs 1984; Lee *et al.* 1985; Arnott *et al.* 1986). The present study uses nutrient manipulations to generate a variety of food regimes to investigate and compare life history responses of *Diaptomus kenai* and *D. leptopus* among a variety of diets. Manipulations of nutrient level and ratio are commonly employed for investigating the role of nutrient limitation in structuring phytoplankton communities both in field (Langeland and Reinertson 1982; Istvanovics *et al.* 1986; Sullivan and McManus 1986; Prepas and Trimbee 1988) and laboratory (Tilman *et al.* 1986; Suttle and Harrison 1988a,b) conditions. In this study, the purpose of fertilization was not to select for specific algal communities, but rather to produce food regimes which differ in species composition and, due to nutrient limitation, chemical composition. Thus, it is not the effects of nutrient manipulations *per se* that are of interest, but rather the effect on copepods of variation in food composition, with nitrogen:phosphorus manipulations being the tool by which these variations are generated.

METHODS

Study Site

The study was conducted in Shirley Lake, a fishless, oligotrophic lake located in the University of British Columbia Malcolm Knapp Research Forest (Fig. 1). The north, east, and south shores of the lake are characterised by humic soils populated with huckelberry, skunk cabbage, yellow cedar, and western red cedar. The western shore is characterised by sandy loam soil, huckelberry, ferns, mosses, lodgepole pine, Douglas fir, and western hemlock. Mean annual precipitaion in the area is typically 280 - 320 cm (Klinka 1976).

"Nutrient" Enclosures

In June 1986, four big-bag enclosures constructed of 4 mil (0.1 mm) transparent polyethylene were suspended from styrofoam-supported wood frames floating at the water surface (Neill 1978, 1981). The enclosures, measuring 1.3 m diameter by 6 m deep, were closed at the lower end to prevent contact with lake sediments and anchored in the center of the lake. Each enclosure was filled by a gasoline-powered pump with 11,000 L of lake water collected in equal volumes from 0.5 m, 2 m, and 4 m depth and filtered through a 76 μm mesh net to remove most zooplankton but not phytoplankton. Zooplankton were collected from the lake using oblique hauls of a 100 μm mesh plankton net. The collected zooplankton were pooled and distributed among the enclosures to give each a zooplankton community similar in concentration and composition to the lake community.

Nutrient Additions

To produce a range of phytoplankton communities, three of the enclosures were fertilized with KH_2PO_4 and NH_4Cl at elemental N:P supply ratios of 10:1, 25:1, and 40:1 (Table I). To avoid confounding desired changes in community composition with large changes in total cell density, nutrients were added at low concentrations. Orthophosphate additions (2 $\mu g \cdot L^{-1}$) were equivalent to orthophosphate levels reported for similar lakes in the Research Forest

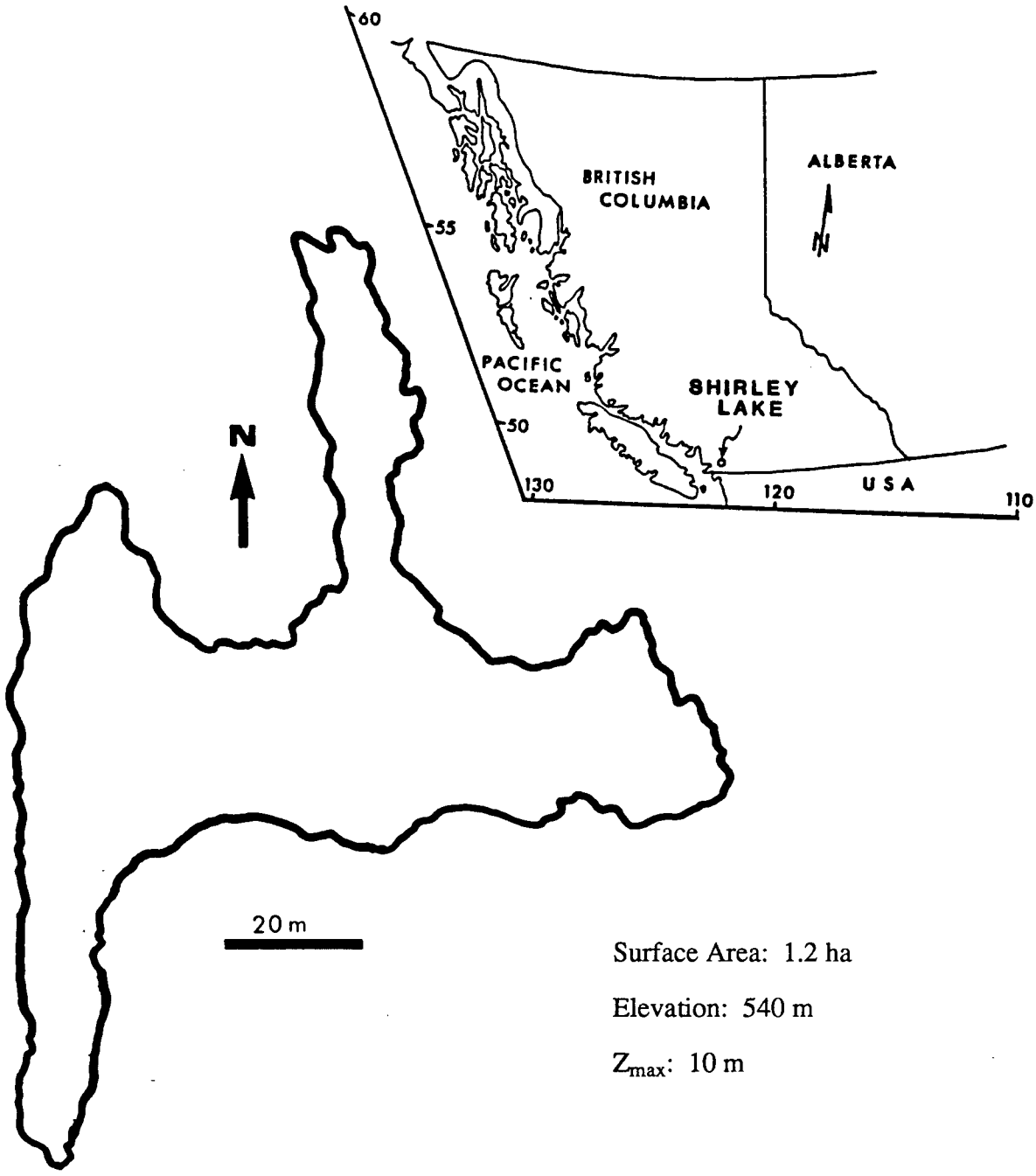


FIGURE 1: Location of Shirley Lake and the U.B.C. Malcolm Knapp Research Forest.

TABLE I: Weekly nutrient additions to the fertilized enclosures.

ENCLOSURE	NH_4Cl (mg)	KH_2PO_4 (mg)
10X	130	33
25X	324	33
40X	518	33
NF	nil	nil

(e.g. Werring 1986). The enclosures were fertilized weekly during the ice-free season (late March to late November) of 1986 and 1987. No nutrients were added during the winter months. After being dissolved in a small volume of water (≈ 20 mL) taken from the enclosure, the nutrients were added to the enclosure and stirred in with an oar. The enclosures were stirred sufficiently to permit mixing throughout their entire depth. The fourth enclosure was stirred but received no nutrient additions. Because the purpose of fertilization was not to produce specific phytoplankton communities but simply to produce different phytoplankton communties, the nutrient manipulations themselves were not replicated.

Sampling Methods

Beginning with the day the enclosures were established, zooplankton and phytoplankton were collected from the lake and enclosures at weekly intervals during the open water period and at monthly intervals during the period of ice cover. Temperatures in the enclosures and the lake were also monitored; no difference was found.

Phytoplankton samples were obtained by pooling equal volumes of water collected with a Van Dorn bottle at 0.25 m, 2 m, and 4 m depth. Samples (250 mL) were preserved in Lugol's solution and settled in graduated cylinders over a 48-h period to reduce the sample volume to 15 mL. This 15 mL sample was then settled in settling chambers for 36 h prior to counting on an inverted microscope at 300X magnification. Although visual inspection did not suggest a bias in the distribution of cells in the chamber, fields were counted in transects from edge to edge in the chamber to minimize effects of the possible bias. The number of fields counted varied with each sample's total cell density, but a sufficient number of fields were counted to ensure adequate representation of the predominant taxa (i.e., until at least 100 cells of each of the more abundant taxa had been recorded). For those samples of low cell density, as much of the entire sample was counted as was possible.

Copepod and zooplankton samples were collected via pooled replicate vertical hauls of a 40 cm diameter plankton net (100 μm mesh). Three hauls from bottom to surface were taken from each enclosure, and five hauls from arbitrary sites in the lake around the enclosures. Samples were preserved in 5% buffered formalin. Taxa were identified using keys of Wilson (1953), Edmondson (1959), and Pennak (1978) and counted using a dissecting microscope at 25X or, when necessary to discern fine structures, an inverted microscope.

Sex and reproductive condition and clutch size of females were recorded for *D. kenai* and *D. leptopus* (Fig. 2). Clutches were removed and egg diameter measured for *D. kenai*. Measurements of cephalothorax length were taken on adult *D. kenai* (Fig. 3). Cephalothorax length and egg diameter were measured to 0.01 mm using an ocular micrometer on a dissecting microscope at 50X magnification.

"Size" Enclosures

Results from the 1986 studies indicated that *D. kenai* in Shirley Lake had two reproductive periods (June and November), with different size categories of females (mean cephalothorax length of 1.04 mm and 1.18 mm, respectively) being involved in each of these reproductive periods. To assess whether these size/reproduction classes were distinct subgroups of the population, I isolated ovigerous *D. kenai* in June and November 1987 into two additional enclosures, which were established in June 1987 in the manner described above. A sufficient number of oblique tows of a 100 μm plankton net were collected to obtain adult *D. kenai* from a volume equivalent to that in the enclosures. Only egg bearing *D. kenai* were added to the enclosures; the remainder of the sample was discarded. One enclosure was seeded in June 1987 with egg-bearing *D. kenai* (termed the "small" enclosure, reflecting the body size of the stocked copepods) and the other was seeded in November 1987 with egg-bearing *D. kenai* (termed the "large" enclosure). Thus, one enclosure contained only the small

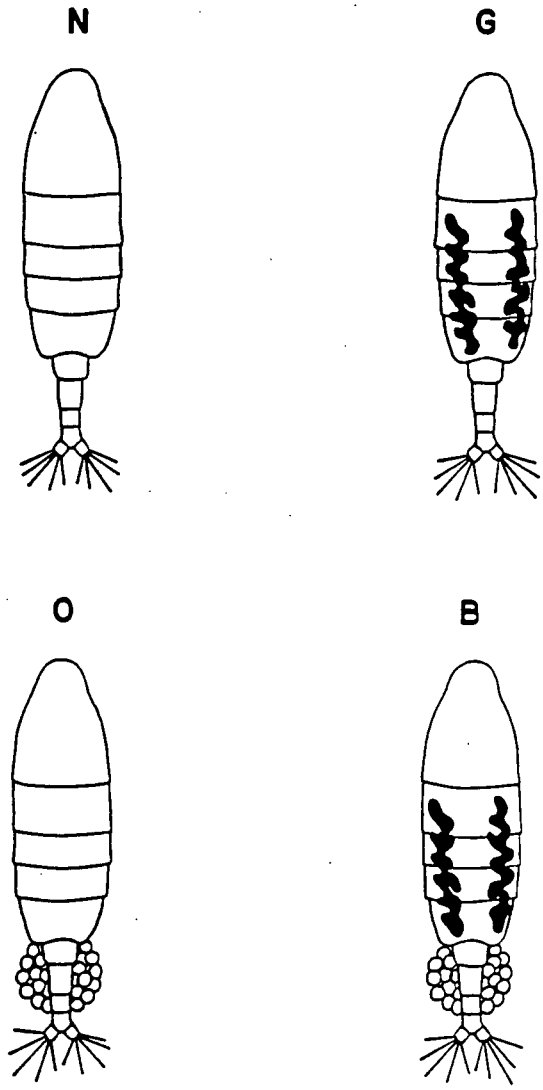


FIGURE 2: Reproductive states of adult female *Diaptomus*. (N: nongravid; G: gravid; O: ovigerous; B: both gravid and ovigerous)

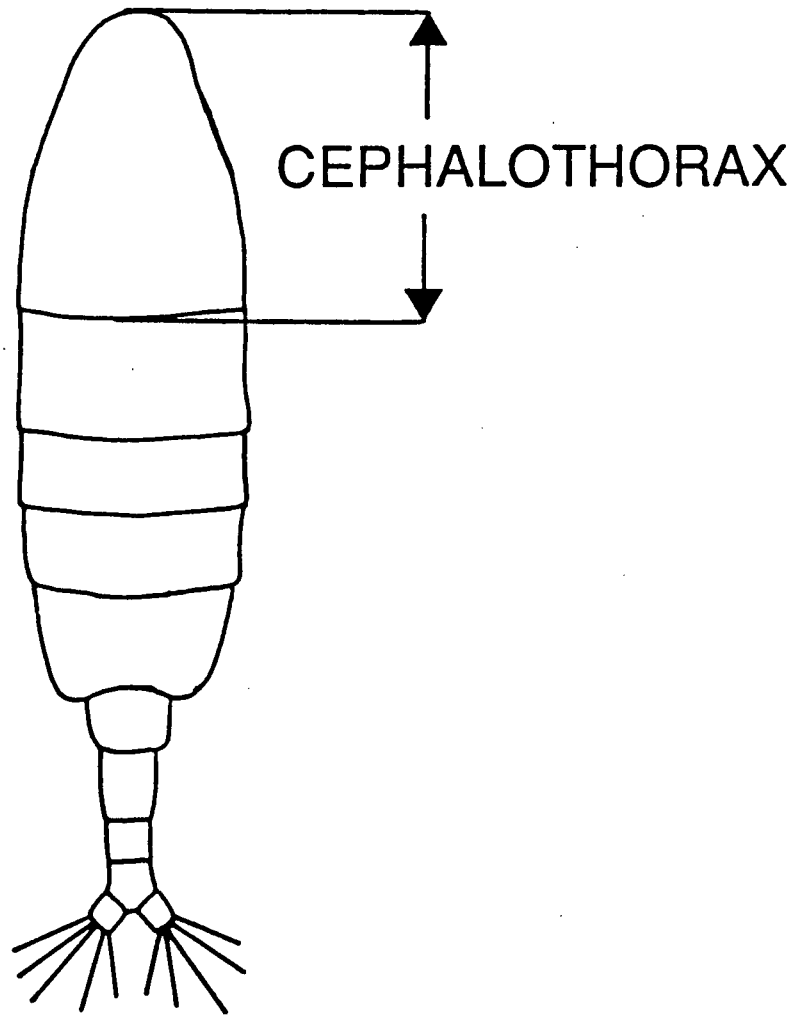


FIGURE 3: Measurement of cephalothorax length in adult *Diaptomus*.

females which produced eggs in the spring and early summer (the "small" enclosure) while the other contained only large females, which produced eggs in the late summer and early fall (the "large" enclosure). Nutrients were added to the enclosures weekly during 1987 and fortnightly during 1988 at the 25:1 N:P ratio described above to maintain a phytoplankton community capable of supporting both size classes (based on results of the 1986 studies).

Copepods (the offspring of the initial females) were collected monthly from the enclosures to measure body size during the period June-November 1988. Clutches were removed, and clutch size and egg diameter recorded. Measurements were made of cephalothorax length, as described above. There was no evidence of egg loss during preservation.

Analyses

Diaptomus kenai population density, *D. leptopus* population density, and total phytoplankton cell densities in the lake and enclosures were analyzed using ANOVA (enclosure x sampling date) without replication (interaction effects pooled with error variance) (Sokal and Rohlf 1968). Although this analysis technique has been applied by others (e.g., Walters *et al.* 1990), I recognize that the results must be interpretted with caution, given the lack of independence in assigning data to the date and enclosure variables and the potential for inaccuracies in calculating the error term. Phytoplankton density data were transformed ($\log_{10}[X+1]$) to meet the assumption of normality. No transformations were necessary for the copepod data.

Phytoplankton Community Structure: To quantify the differences in food regimes in each enclosure and the lake, 1986 phytoplankton community composition was compared among the four enclosures and the lake using discriminant analysis (i.e. canonical variate analysis) of the absolute density of each taxa. Discriminant analysis emphasizes differences among groups to identify those taxa which are most significant in distinguishing among the communities. Because this technique provides an ordination of the group means adjusted for

within-group variation, it is appropriate for comparing among samples collected over time (Digby and Kempton 1987). Wilks' lambda is the proportion of the total variability that is not explained by between group differences (in this case, differences between enclosure communities). Using a forward entry algorithm for selection of variables to minimize Wilks' lambda, identifies the variables (in this case, genera) which are most important in separating the groups (Norusis 1988). Four genera (*Mallomonas*, *Dinobryon*, *Staurastrum*, *Scenedesmus*) never exceeded one percent of the total cell number on any date in any enclosure and were excluded from analyses.

Body Length: Variation in adult *Diaptomus kenai* body length was analyzed with a fixed effects, nonorthogonal (due to unequal sample sizes) ANOVA to test for differences among the lake and enclosures and between males and females. The data met the assumptions of homoscedasticity and all data groups except the lake were normally distributed; therefore, the data were not transformed. Body size of *D. leptopus* was not analyzed.

Egg Diameter and Clutch Size: The dependence of *D. kenai* and *D. leptopus* clutch size on food density was estimated with regression analysis and variation attributable to enclosure effects was tested with one-way ANOVA. Correlation analysis was used to estimate the association between variation in *D. kenai* clutch size and variation in egg diameter and adult female body size. Because clutch size is strongly associated with body length, analysis of covariance isolated the effect of the enclosure treatments from the effects of body size on clutch size. Clutch size data from the lake and the "nutrient" enclosures were log-transformed to meet the assumptions of the model. "Large" and "small" enclosure effects on clutch size were tested with one-way analysis of variance with least significant differences (LSD) *a priori* tests for significantly different pairs. No transformations were necessary.

All analyses were performed using the statistical packages SPSS-PC+ and SPSS-PC+ Advanced Statistics (Norusis 1986, 1988).

RESULTS

Phytoplankton Community Structure

Phytoplankton population density changed over time in a pattern consistent among all enclsoures and the lake, with peak densities reached in mid July. Two-way ANOVA (date x enclosure) indicated significant date and enclosure effects on total phytoplankton cell density (Table II). Lake phytoplankton were markedly less dense than in the enclosures (≈200 cells·mL^{-1} compared to as many as 10^4 cells·mL^{-1}). Of the four enclosures, the 10X and NF enclosures had the lowest cell density (maximum of 10^3 and 4 x 10^3 cells·mL^{-1}, respectively) followed by the 25X (maximum of 5.5 x 10^3 cells·mL^{-1}) and 40X (maximum of 10^4 cells·mL^{-1}) enclosures. All four enclosures had marked increases in cell density within one week of setting up the enclosures, although cell density in the 10X and NF enclosures declined steadily over the course of the summer to cell densities similar to lake density. The 25X and 40X enclosures maintained high cell densities until mid-October, at which time cell numbers declined sharply to lake levels.

While eight phytoplankton taxa were common in the lake and enclosure communities (Fig. 4), discriminant analysis with minimization of Wilks' lambda identified three genera (*Crugigenia*, *Chlamydomonas*, and *Fragillaria*) as important in distinguishing among the lake and enclosure communities, based upon cell densities (Table III). *Chlamydomonas* and cryptomonads, generally recognized as an important food source for zooplankton (Reynolds 1984), were a major component of the lake phytoplankton community, where they together accounted for as much as 50 percent of total cell numbers. Neither species was abundant in the enclosures, although late summer densities (as percent of total phytoplankton density) of *Chlamydomonas* approached lake values in all enclosures except the 40X enclosure and cryptomonads bloomed briefly in the 10X enclosure in October.

TABLE II: Analysis of total phytoplankton cell density in the lake and enclosures.

ANOVA TABLE

Source	Sum Squares	DF	F	P <
Enclosure	3.032	4	6.153	0.01
Date	4.173	4	8.467	0.001
Residual	1.725	14		

FIGURE 4: Phytoplankton composition of the lake and enclosures in 1986, expressed as percent of total phytoplankton community. (CHLAMY: *Chlamydomonas*; CRUCIG: *Crucigenia*; SELENA: *Selenastrum*; ANKIST: *Ankistrodesmus*; FRAGIL: *Fragillaria*; COSMAR: *Cosmarium*; MONADS: cryptomonads; CHROOC: *Chroomonas*)

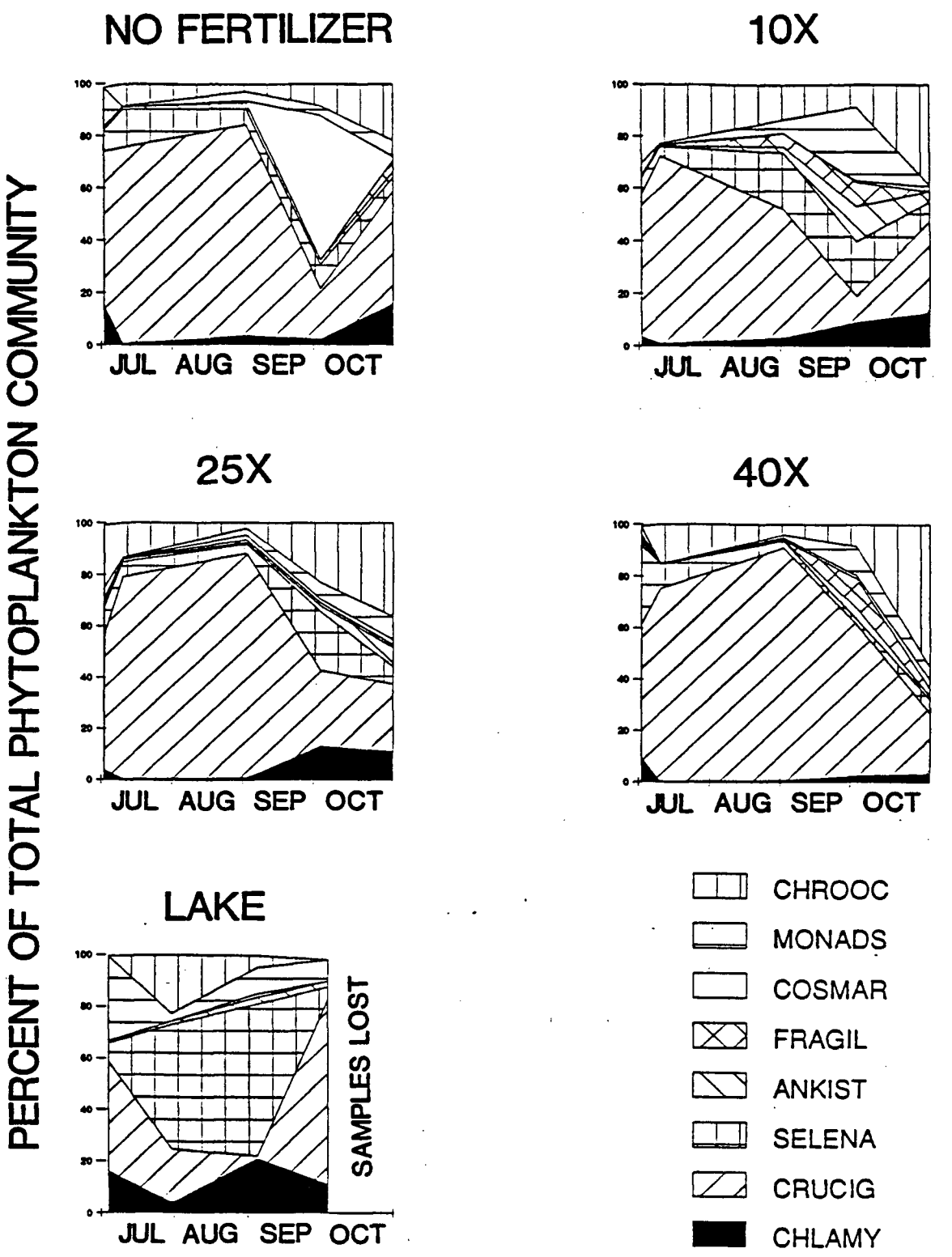

NO FERTILIZER
10X
25X
40X
LAKE
PERCENT OF TOTAL PHYTOPLANKTON COMMUNITY
JUL AUG SEP OCT
SAMPLES LOST
CHROOC
MONADS
COSMAR
FRAGIL
ANKIST
SELENA
CRUCIG
CHLAMY

TABLE III: Summary table of discriminate analysis of phytoplankton species composition.

CANONICAL DISCRIMINANT FUNCTIONS

Function	Eigen Value	% of Variance	Cumulative Variance	Canonical Correlation	After Function	Wilks' Lambda	Chi-Square	DF	$P <$
					0	0.292	22.16	12	0.05
1	0.848	52.7	52.7	0.678	1	0.540	11.10	6	0.10
2	0.615	38.2	90.8	0.617	2	0.871	2.48	2	0.29
3	0.148	9.2	100.0	0.3587					

STANDARDIZED CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS

Taxa	Func. 1	Func. 2	Func. 3
Crucigenia	0.4504	0.8635	0.3897
Chlamydomonas	0.8526	0.0586	-0.5590
Fragilaria	0.3877	-0.7539	0.5815

Copepod Population Dynamics

Seasonal changes in population density of *Diaptomus kenai* and *D. leptopus* are presented in Figures 5 and 6, respectively. Two-way ANOVA indicated significant effects of date and enclosure on population density for both species (Tables IV and V).

Adult *D. kenai* numbers peaked in late summer (July/August) and declined steadily until late December, after which no *D. kenai* remained in the water column. Nauplii appeared in late winter and by late May the population consisted almost entirely of adults. Population density in the 25X and NF enclosures equalled or exceeded lake densities. The 10X enclosure was similar to lake density, while, by mid-summer of 1987, the population in the 40X enclosure was reduced to less than 0.1 individual·L^{-1}.

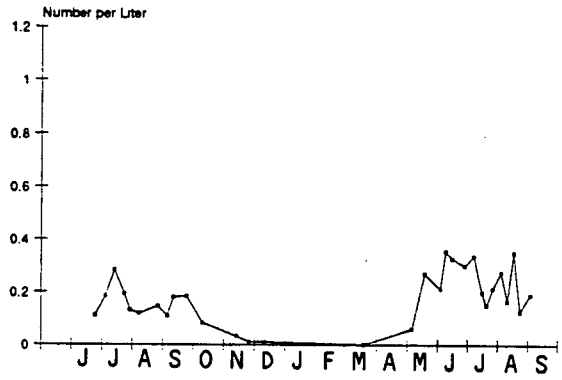
Adult *D. leptopus* population density peaked in October, declining steadily through the spring to remain low until the following September, at which time population density began to increase. Population density of *D. leptopus* was substantially reduced in all the enclosures compared with the lake, particularly in the 40X enclosure, in which none remained after early June of 1987.

Diaptomus kenai Size Distribution

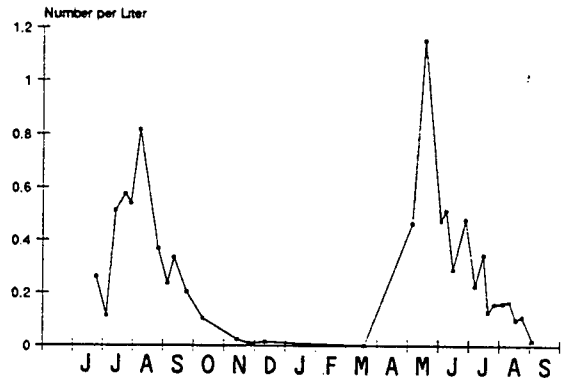
"Nutrient" Enclosures: Cephalothorax length of adult *Diaptomus kenai* collected in 1987 and 1988 varied significantly both between sexes and among the lake and enclosures (Fig. 7; Table VI). Mean length of copepods in the 10X enclosure was greater than in the other enclosures (males = 1.10 mm, females = 1.25 mm), while the No Fertilizer population had the smallest mean size (males = 0.98 mm, females = 1.01). There were significant interactions between sex and enclosure, indicating the effect of the enclosure on body size differed between males and females. There was a more pronounced enclosure effect on mean cephalothorax length for females than for males. Male cephalothorax length varied little among the enclosures, ranging from 1.01 to 1.02 mm in four out of the five enclosures,

FIGURE 5: *Diaptomus kenai* population density in the lake and enclosures during 1986 and 1987.

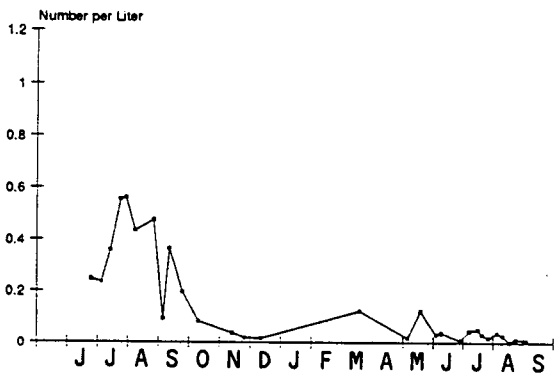
10X



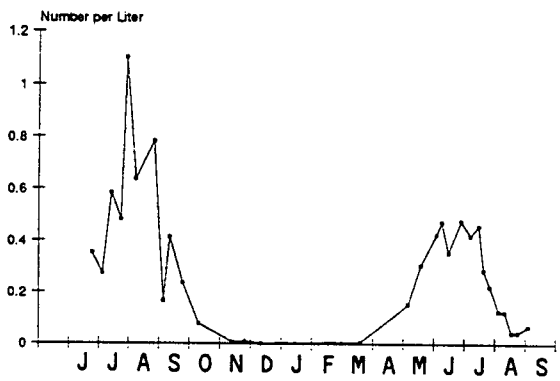
25X



40X



N.F.



LAKE

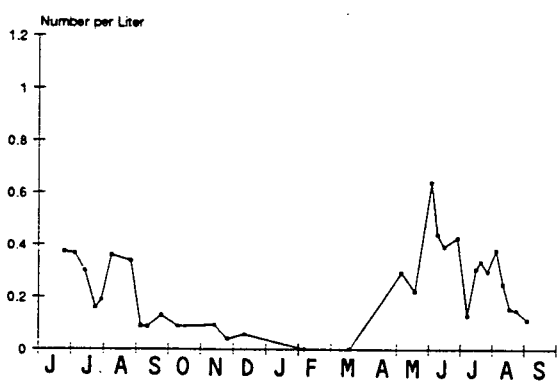
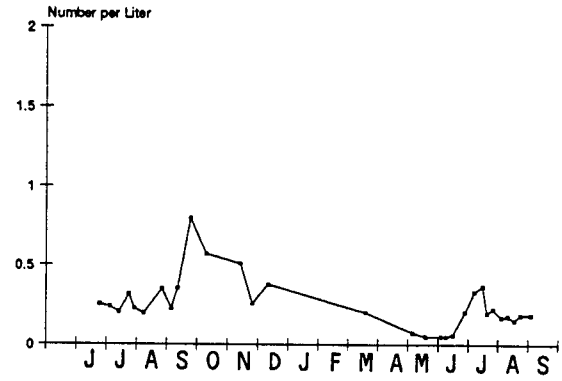
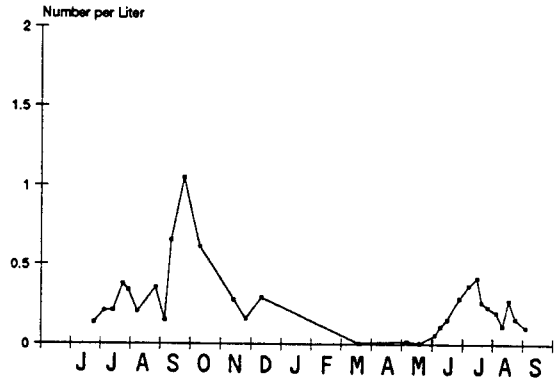


FIGURE 6: *Diaptomus leptopus* population density in the lake and enclosures during 1986 and 1987.

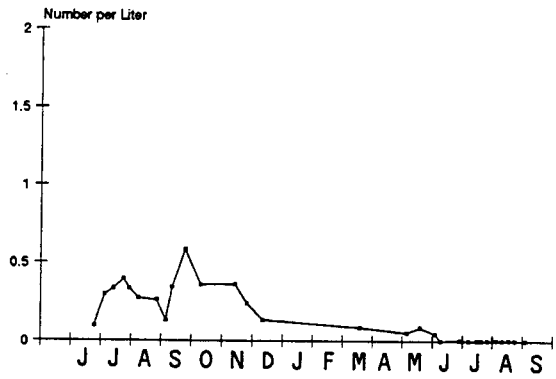
10X



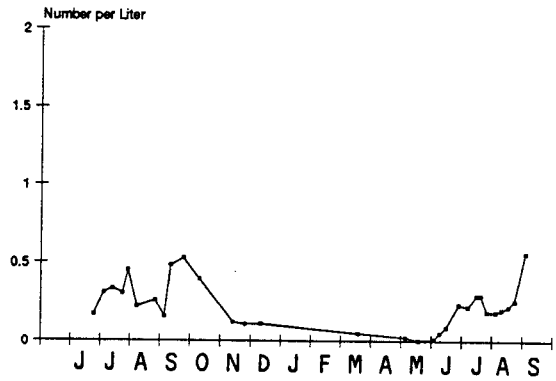
25X



40X



N.F.



LAKE

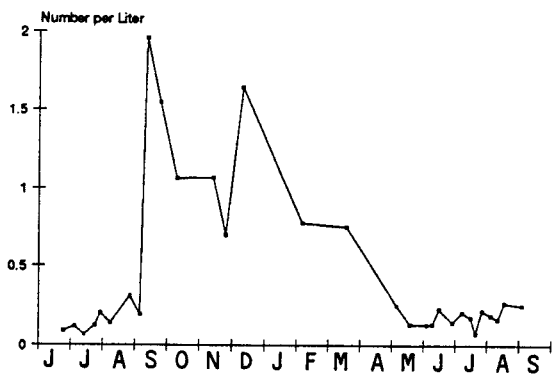


TABLE IV: Analysis of *Diaptomus kenai* population density in a) 1986 and b) 1987.

ANOVA TABLE

a) 1986

Source	Sum Squares	DF	F	*P* <
Enclosure	0.42	3	5.596	0.002
Date	1.61	6	10.730	0.001
Residual	1.51	46		

b) 1987

Source	Sum Squares	DF	F	*P* <
Enclosure	0.56	3	7.992	0.001
Date	0.57	5	4.913	0.001
Residual	1.26	54		

TABLE V: Analysis of *Diaptomus leptopus* population density in a) 1986 and b) 1987.

ANOVA TABLE

a) 1986

Source	Sum Squares	DF	F	*P* <	R
Model	0.59	9	2.548	0.02	0.33
Enclosure	0.06	3	0.803	0.50	
Date	0.53	6	3.421	0.01	
Residual	1.18	46			

b) 1987

Source	Sum Squares	DF	F	*P* <	R
Model	0.51	8	7.506	0.001	0.53
Enclosure	0.28	3	11.059	0.001	
Date	0.24	5	5.624	0.001	
Residual	0.46	54			

FIGURE 7: Frequency distribution of adult female *Diaptomus kenai* cephalothorax length in the lake and enclosures.

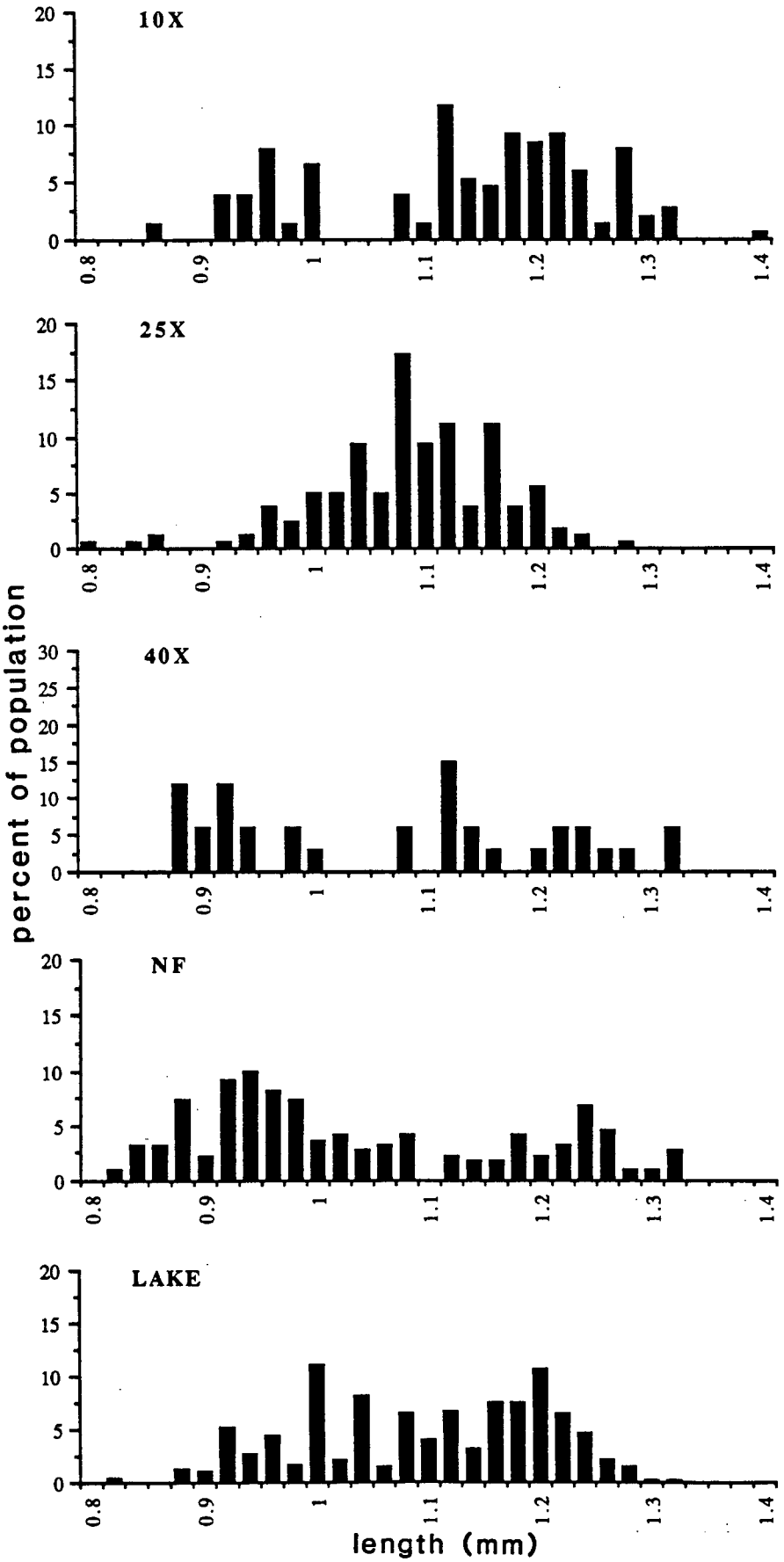

10X
25X
40X
NF
LAKE
percent of population
length (mm)
0.8
0.9
1
1.1
1.2
1.3
1.4

TABLE VI: Analysis of *Diaptomus kenai* body length (lake and enclosures).

ANOVA TABLE

Source	Sum Squares	DF	F	P <
Sex	0.751	1	72.889	0.001
Enclosure	1.308	4	31.730	0.001
Interaction	0.443	4	10.740	0.001
Error	8.782	852		

while female cephalothorax length ranged from 0.98 mm (NF enclosure) to 1.25 mm (10X enclosure). Only in the 10X enclosure did male body size differ significantly from what was observed in the lake, while there was significant variation in female cephalothorax length among the lake and enclosures. It would appear, therefore, that males are less responsive to the factors affecting body size than are females.

"Size" Enclosures: Cephalothorax length of adult *D. kenai* collected in 1988 varied significantly between the enclosures in a manner consistent with the size distributions of the females introduced to the enclosures the previous year, suggesting population size structure of the offspring reflected maternal body size (Fig. 8, Table VII). Copepods hatched from "large" females developed into larger copepods than those hatched from "small" females. There was no significant interaction between sex and enclosure in affecting body size; both sexes were equally different in the two enclosures.

Reproduction in the Lake and "Nutrient" Enclosures

Adult *D. leptopus* were present in Shirley Lake year round, but reproduction was limited to the period between April and August, when population density was at its lowest point (Fig. 9). Reproduction peaked in July with 100% of the female population reproducing in all enclosures except the 40X enclosure. In that enclosure there was a decline in the percentage of the females reproducing never exceeded 50% and there was a reduction in the duration of the reproductive period, culminating in total die-off of the population by the end of June.

There was no evidence of subitaneous egg production by *D. leptopus*; all females observed with eggs carried resting eggs. Clutch size varied considerably among the enclosures (Fig. 10). One-way ANOVA indicated significant enclosure effects on clutch size ($F = 11.01$; $p < 0.001$), but regression analysis indicated that clutch size was not signficantly dependent upon maximum phytoplankton density in the enclosures ($R^2 << 0.01$; $p > 0.05$).

FIGURE 8: Frequency distribution of adult female *Diaptomus kenai* cephalothorax length in the "size" enclosures. (Averaged from samples collected in 1988)

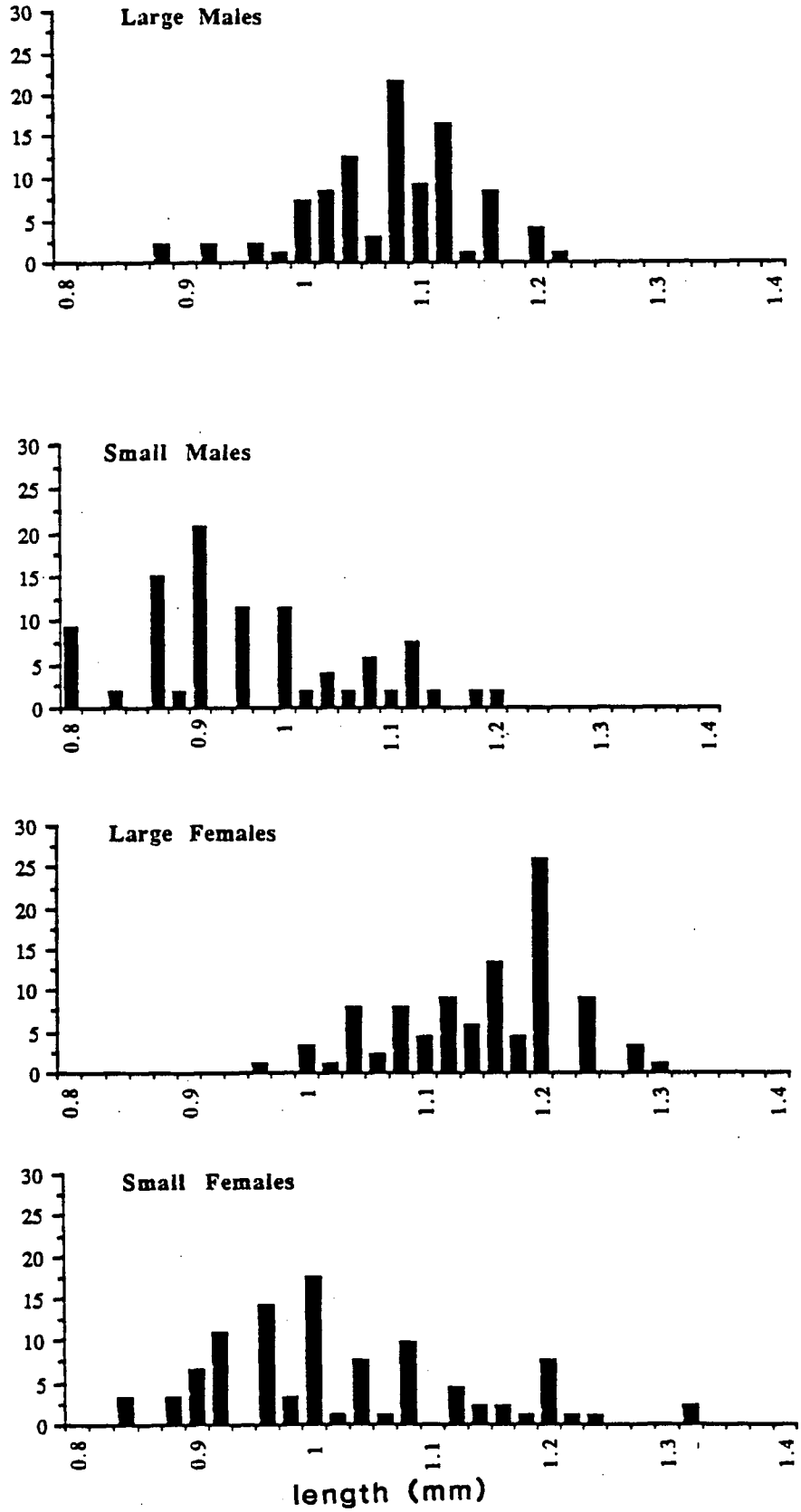

Large Males
Small Males
Large Females
Small Females
30
25
20
15
10
5
0
0.8
0.9
1
1.1
1.2
1.3
1.4
length (mm)

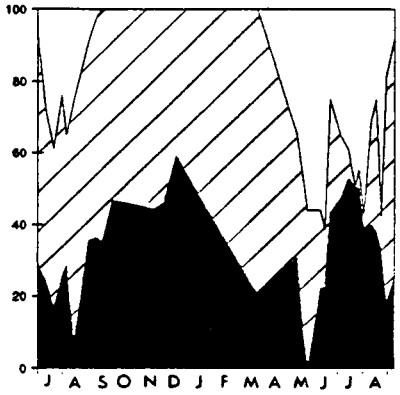
TABLE VII: Analysis of *Diaptomus kenai* body length in the large and small enclosures.

ANOVA TABLE

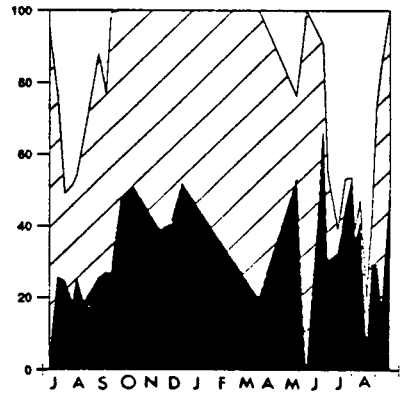
Source	Sum Squares	DF	F	P <	R
Model	1.347	3	58.077	0.001	0.35
Sex	0.363	1	46.992	0.001	
Enclosure	1.147	1	148.256	0.001	
Interaction	0.006	1	0.743	0.39	
Residual	2.529	327			

FIGURE 9: Composition of *Diaptomus leptopus* populations in the lake and enclosures during 1986 and 1987. (Population disappeared completely from the 40X enclosure in June 1987.)

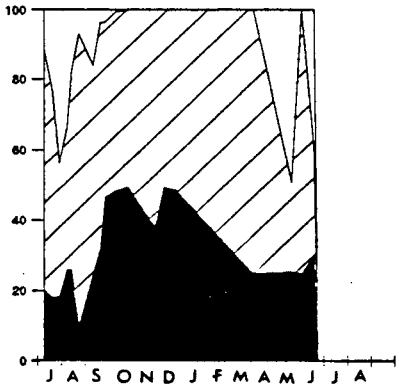
10X



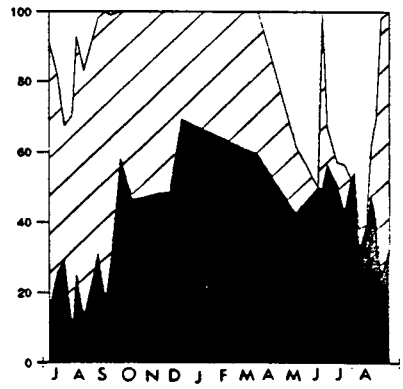
25X



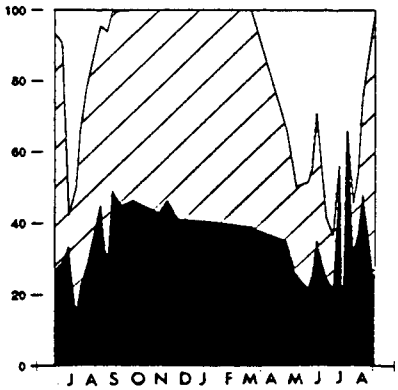
40X



NO FERTILIZER



LAKE



MALE

NONGRAVID

REPRODUCTIVE

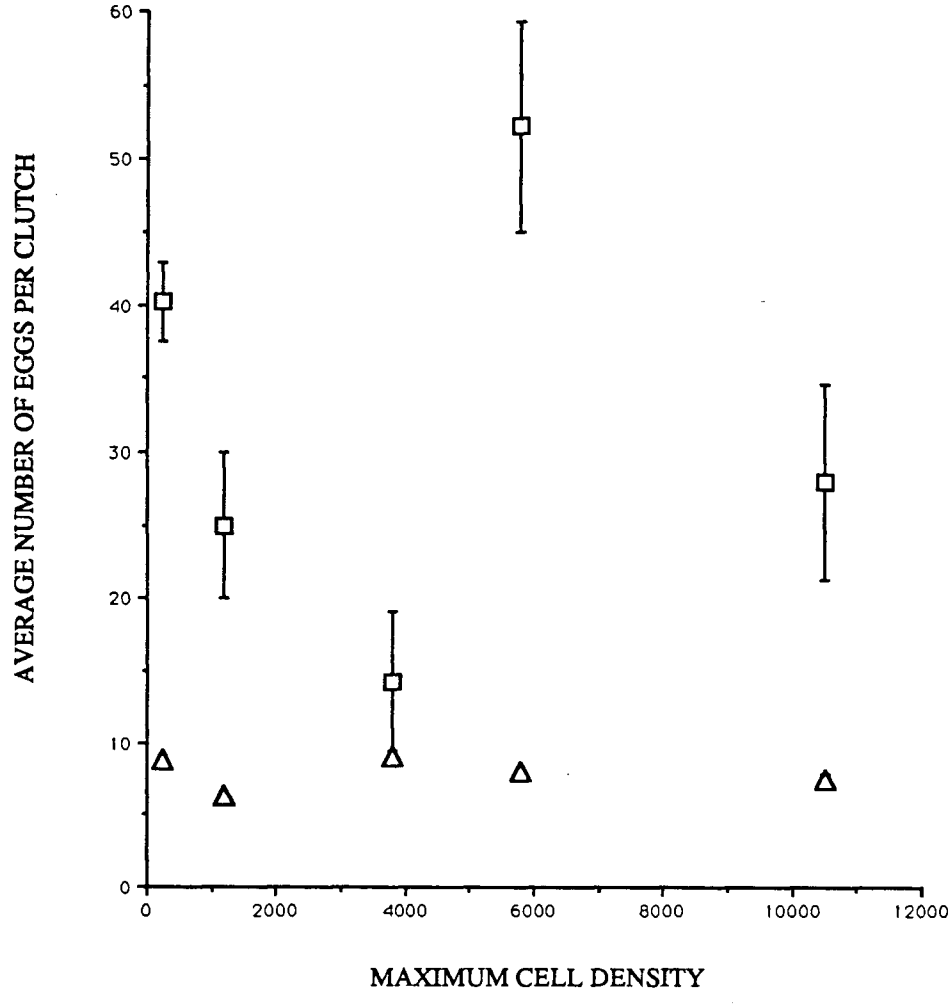


FIGURE 10: *Diaptomus kenai* (squares) and *D. leptopus* (triangles) clutch size versus maximum cell density (lake and enclosures). Standard error (where greater than size of symbol) indicated by vertical bars.

Adult *D. kenai* in Shirley Lake were characterized by two periods of reproduction, one occurring in May to July and the other in October to December (Fig. 11). Approximately 25-30% of the females present in the spring population reproduced at that time, while 50-100% of the females present in the fall reproduced during that period. All females produced resting eggs; there were no subitaneous eggs. The 10X and 25X enclosures' *D. kenai* populations had reproductive patterns similar to what was observed in the lake. *D. kenai* in the 40X and NF enclosures had reduced reproduction, as reflected in the short durations of the spring and fall reproductive periods, respectively.

The mean size of females reproducing in the lake during the late spring was compared to that of females reproducing in the fall with Student's t-test. Because the variances of the two groups were not equal, a separate-variance t-test (Sokal and Rohlf 1969) was used. Females reproducing in the late spring are significantly smaller than females reproducing in the fall (Table VIII).

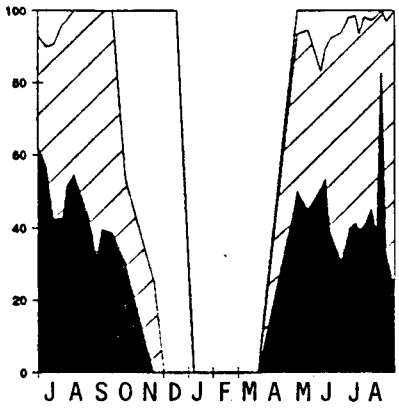
Analysis of covariance controlling for the effects of body size indicated that enclosure had a significant effect on *D. kenai* clutch size (Table IXa). Regression analysis indicated that clutch size was not signficantly dependent upon maximum phytoplankton density (R^2 = 0.0196; $p > 0.05$). Clutches produced by females in the 10X and 25X enclosures had three to four times as many eggs as those produced by females in either the lake or the NF enclosure. There was no correlation between clutch size and egg diameter, but clutch size and body size were strongly correlated ($R = 0.572$; $p < 0.001$) (Fig. 12).

Reproduction in the "Size" Enclosures

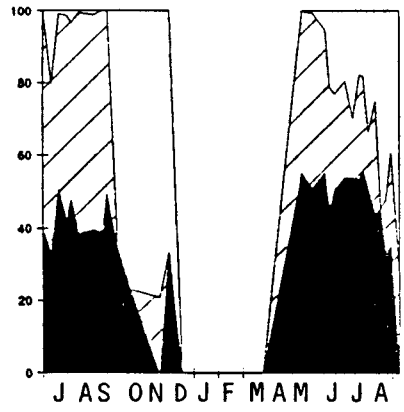
Female *D. kenai* in the "large" enclosure produced clutches that were 50% larger than those produced by females in the "small" enclosure. There was no correlation between clutch size and egg diameter, but clutch size and body size were strongly correlated ($R = 0.506$; $p <$

FIGURE 11: Composition of *Diaptomus kenai* populations in the lake and enclosures during 1986 and 1987. (Gaps during the winter months indicate no adults were present)

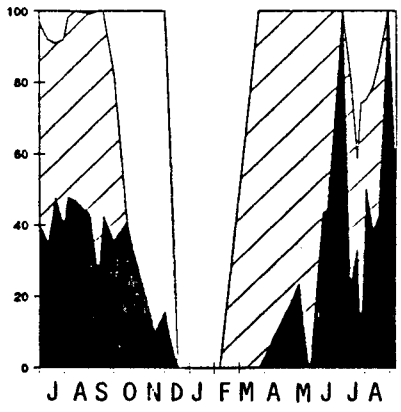
10X



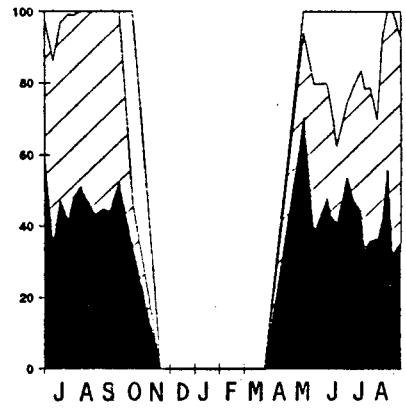
25X



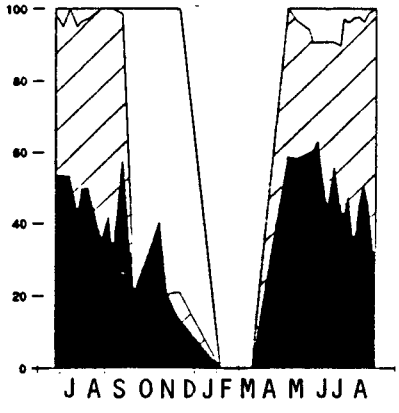
40X



NO FERTILIZER



LAKE



MALE

NONGRAVID

REPRODUCTIVE

TABLE VIII: Results of separate variance t-test comparison of size of reproducing *Diaptomus kenai* in the spring and summer versus those reproducing in the fall.

Reproductive Season	Number of Cases	Mean (mm)	Standard Deviation	Standard Error
Spring/Summer	139	1.04	0.079	0.007
Fall	99	1.18	0.064	0.006

Separate Variance t-Test

t Value	Degrees of Freedom	2-Tail Probability
-15.89	231.67	0.001

TABLE IX: Analysis of *Diaptomus kenai* clutch size for a) the lake and "nutrient" enclosures and b) the "size" enclosures, with main (enclosure) and covariate (body size and egg diameter) effects.

ANOVA TABLE

a) lake and "nutrient" enclosures

Source	Sum Squares	DF	F	*P* <
Model	3.884	5	2.548	0.02
Enclosure	1.922	3	0.803	0.05
Diameter	0.000	1	0.001	0.973
Length	1.674	1	37.200	0.001
Residual	3.330	74		

b) "size" enclosures

Source	Sum Squares	DF	F	*P* <
Model	393.745	3	3.135	0.05
Enclosure	1.947	3	0.047	0.83
Diameter	19.495	1	0.466	0.50
Length	376.128	1	8.985	0.005
Residual	1423.334	34		

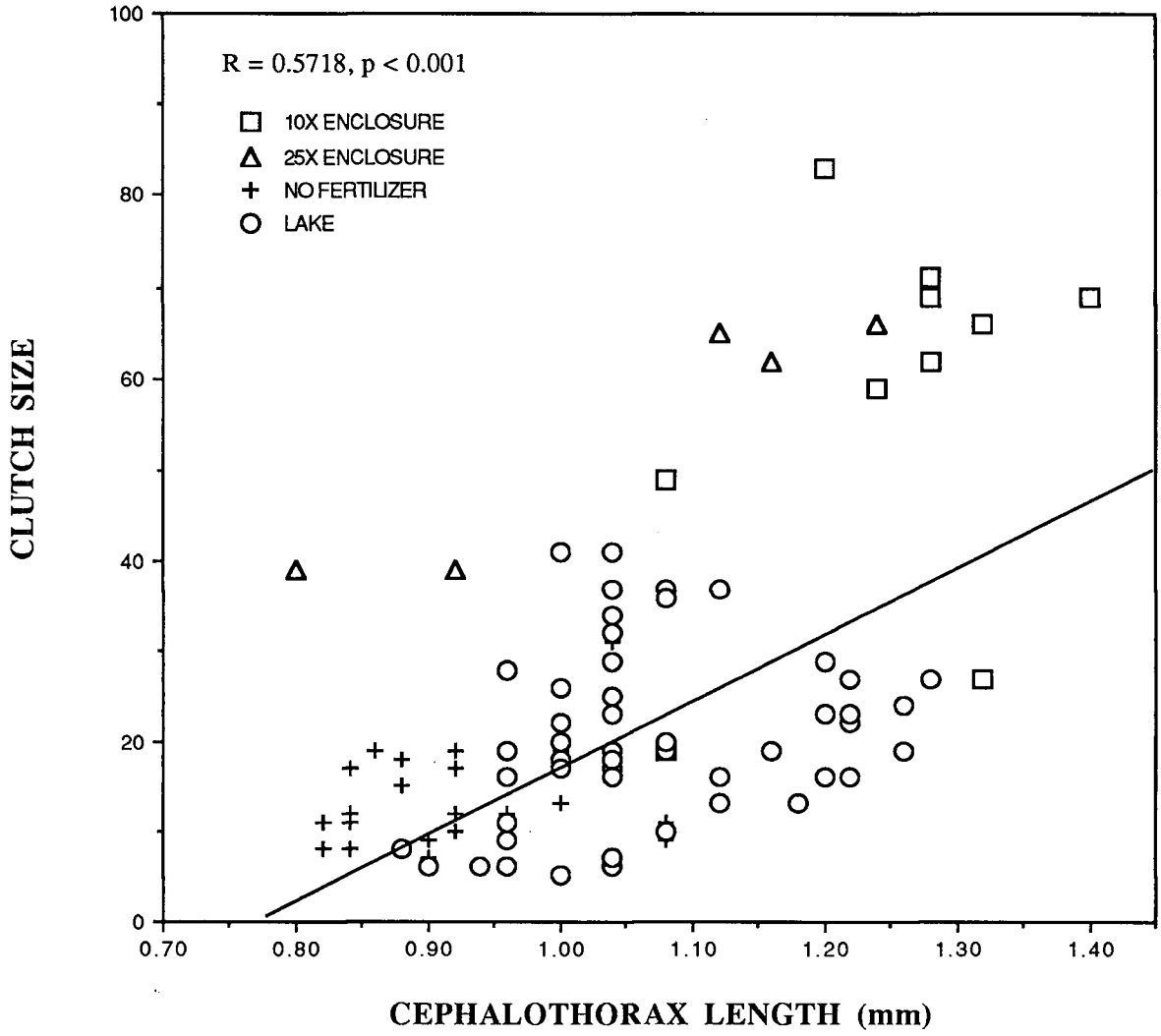


FIGURE 12: Clutch size versus cephalothorax length of *Diaptomus kenai* from the lake and enclosures. Diagonal line (fitted by eye) approximates the linear association of clutch size and cephalothorax length in the lake population.

0.001) (Fig. 13). Analysis of covariance controlling for the effects of body size and egg diameter indicated that there was no significant enclosure effect on clutch size for the "large" and "small" enclosures (Table IXb).

DISCUSSION

During the first year, there was a significant enclosure effect on clutch size (to which I shall refer as the reproductive time scale) for both *Diaptomus kenai* and *D. leptopus*, suggesting that an attribute of the enclosure (for example, food quality or food concentration) was affecting fecundity. Other studies have shown that such an effect can be realized within days of a change in the food supply (e.g., Williamson *et al.* 1985). Laboratory studies of marine (Checkley 1980a; Abou Debs and Nival 1983; Runge 1984; Smith and Lane 1985; Arnott *et al.* 1986) and freshwater (Woodward and White 1981; Elmore 1982; Williamson *et al.* 1985) copepods have shown egg production to be severely limited by low food concentrations. Because these studies have generally involved manipulating the density of a unialgal food supply, it is difficult to apply their results to field conditions, although there is evidence of maximum fecundity in nature coinciding with periods of phytoplankton blooms (Edmondson *et al.* 1962; Durbin *et al.* 1983; Peterson 1985). However, the enclosure effect observed in this study could not be attributed to the significant differences in cell density or cell volume among the lake and enclosures. In fact, the enclosure with the highest cell concentration had the lowest clutch size for both *D. kenai* and *D. leptopus*. A similar detrimental effect of high cell concentrations has also been observed in studies of calanoid nauplii survival (Williamson *et al.* 1985), with decreased survival at the highest cell densities.

It is also possible that the observed variation in clutch size is a response to food composition, given the differences in phytoplankton community structure observed among the lake and

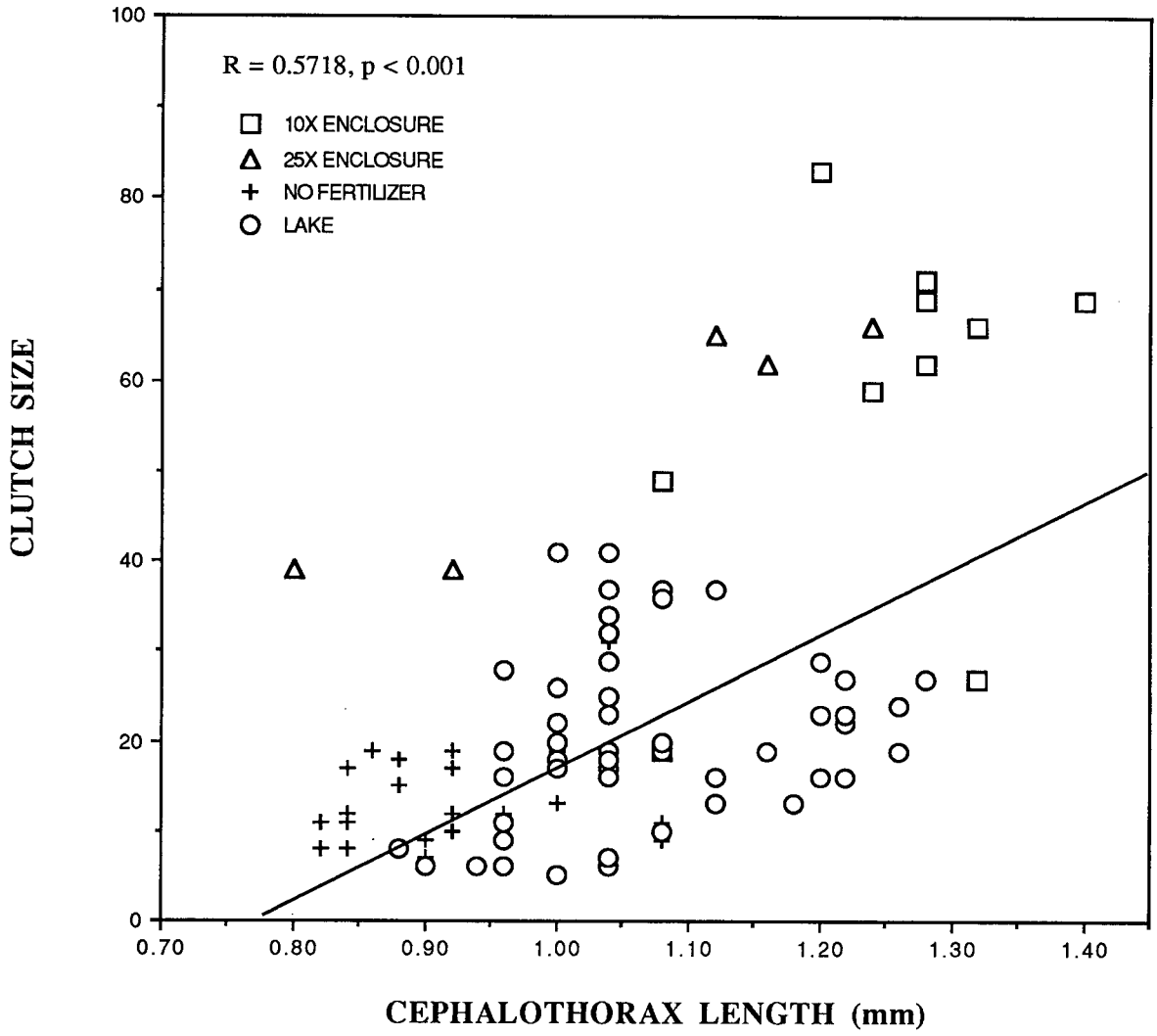


FIGURE 13: Clutch size versus cephalothorax length of *Diaptomus kenai* from the "large" and "small" enclosures.

enclosures. Thus the response may be have resulted from variation in food quality than to food quantity. Laboratory studies have demonstrated that clutch size can be affected by variations in food quality, whether food quality is defined by chemical composition of an artificial diet (Gaudy and Guérin 1977) or species composition (Cowgill *et al.* 1985; Lee *et al.* 1985; Arnott *et al.* 1986; Williamson and Butler 1986).

The potential role of food quality is further supported by the changes observed on what I term the demographic time scale, the changes in copepod population density which can occur over weeks (when the change is due to mortality of the existing population) or seasons or years (when the change is due to reproductive success). As with clutch size, although there is a significant enclosure effect on copepod population density, there is no correlation between population density and cell density, again suggesting that some attribute of the food besides its density may be important in determining copepod population density. The fact that the change in population density was evident during the first year of the study suggests that mortality rates were affected, given that the repercussions of an effect on production and viability of resting eggs would not be realized until the second year. The off-set population peaks, with *D. kenai* reaching maximum population density around July and *D. leptopus* in October, are similar to what has been observed for coexisting cladocerans (Culver 1980) and copepods (Evans *et al.* 1986; Moraitou-Apostolopoulou *et al.* 1986; Threlkeld and Dirnberger 1986).

Not only were there significant enclosure effects on clutch size and population density, but there was also a marked difference in body size among the lake and enclosure *Diaptomus kenai* populations. Analysis of size structure within the Shirley Lake population indicated a bimodal distribution of two co-occurring size classes. In 1987, the size structure of the *D. kenai* populations ranged among the enclosures from unimodal distributions consisting predominantly of either large (10X) or small (NF) sized copepods to distributions similar to

that observed in the lake (25X). Variation in body size within a copepod population is not unusual and is generally attributed to seasonal phenomena. For example, within populations of multivoltine copepod species, cohorts that hatch and develop at cold temperatures tend to grow into larger adults than cohorts that hatch and develop at warmer temperatures (McLaren 1965; Patalas and Patalas 1978; Allan and Goulden 1980; Evans 1981; Head and Harris 1985; Crawford and Daborn 1986; Myers and Runge 1986; Klein Breteler and Gonzalez 1988; McLaren *et al.* 1988). However, it is extremely rare to find two size classes occurring simultaneously except when attributed to sexual size-dimorphism (e.g. Maly 1973; Grigg *et al.* 1987). The observed variation in *D. kenai* body size among the lake and enclosures does not appear to be a growth response to physical environmental factors. For example, there was no discernable difference among the lake and enclosures in such physical attributes as temperature, thermal stratification, or day length that might contribute to differences in growth rate or development rate.

Because the differences in size structure cannot be readily attributed to physical characteristics of the enclosures, it is possible that size structure is a reflection of some biological attribute of the enclosures, such as food quality or quantity, affecting perhaps differential survival or growth patterns. Effects of food quality and quantity on copepod growth and mortality have been well documented. Naupliar survival can be affected by food quality (Herzig *et al.* 1980; Arnott *et al.* 1986; Huntley *et al.* 1987) and quantity (Arnott *et al.* 1986). Development rate of nauplii and copepodites can also vary according to the quality (Lee *et al.* 1985; Huntley *et al.* 1987) as well as the quantity (Kimmerer and McKinnon 1987) of the food, resulting in different sizes at maturity. Pessotti *et al.* (1986) observed four to five size classes in field populations of adult *Temora longicornis*, with the smaller sizes being distributed in the nearshore populations and the larger sizes in the offshore populations. Their results indicated that food abundance, not temperature, was the dominant factor influencing body size. However, Wyngaard (1986a) observed that offspring of *Mesocyclops edax* taken from populations characterised by either large or small body size grew to a size

reflecting that of their parent population regardless of the food or temperature conditions under which they were reared. Her results suggest that body size can also be strongly influenced by a heritable genetic component.

If the observed variation in *D. kenai* body size reflects only plastic growth responses to environmental conditions, the size at maturity of cohorts produced by large and small adults should be similar when reared under similar environmental conditions. However, if size is a phenotypically invariant morphological trait, small adults should produce offspring that are significantly smaller at maturity than offspring produced by large adults. When reared separately so cohorts could be distinguished, small adults did tend to produce offspring that grew into small adults while large adults produced offspring that grew into large adults. It would appear, therefore, that size in this population might also be a heritable genetic attribute.

In the lake and nutrient enclosures, there was a significant correlation between body size and clutch size, similar to what has been reported for numerous other vertebrate and invertebrate species, including mayflies (Sweeney *et al.* 1986), guppies (Reznick 1983), marine copepods (Crawford and Daborn 1986), and freshwater copepods (Maly 1983). The correlation between body size and clutch size was modified, however, by enclosure effects, suggesting again that some attribute of the food was influencing reproduction. However, in the "large" and "small" enclosures, which were treated with identical nutrients, there was no significant enclosure effect and observed variation in clutch size was attributed to body size. Thus it would appear that the change in body size is a demographic time-scale event, involving a combination of differential survival and reproductive success which affects size structure on a time scale of seasons and years.

These results suggest that body size in *D. kenai* has a strong genetic component and is not solely a response to some environmental factor such as temperature or food. However, the

fact that the four nutrient enclosures produced different adult sizes, coupled with the pronounced effect of the enclosures on *D. kenai* and *D. leptopus* population density and clutch size, suggests that the different size classes responded differently to the conditions present in each enclosure. As the most obvious difference among these enclosures lay in phytoplankton community structure, it is strongly suggestive that some aspect of the food regime, either quality or quantity, influences survival and reproduction in this animal. The next chapter reports on the effects of food quality and quantity on energy storage of copepods in the enclosures and in the laboratory.

Chapter Three

LIPID STORAGE IN COPEPODS: EFFECTS OF INTER- AND INTRASPECIFIC VARIATION IN FOOD QUALITY

Changes in lake nutrient supplies and phytoplankton community structure represent variation in both the quantity and quality of food available to grazing zooplankton. These changes can occur over time scales ranging from minutes, such as microscale nutrient patches excreted by zooplankton (Lehman and Scavia 1982), to hours or days, such as experienced during lake turnover, to seasons, such as periods between lake turnover events (Wetzel 1982). Because zooplankton can be highly susceptible to starvation mortality (Threlkeld 1976; Williamson *et al.* 1985), individuals capable of accumulating and utilizing a stored energy source could potentially enhance their survival, leading consequently to enhanced growth and reproduction.

The importance of lipid stores (both amount and type) depends upon the nature of the variation in the food regime (magnitude and frequency) as well as the sensitivity of the organism to the variation. An organism with a life cycle shorter than the time scale of the variation in food would most likely not need a source of stored energy. On the other hand, an organism that is likely to experience episodes of food limitation during its lifetime would be more likely to store lipids. For example, arctic and deep sea copepods, which live in habitats characterized by low food levels, occasionally experience periods of high food abundance. During these periods, they accumulate wax esters which are utilized only under starvation conditions (Lee *et al.* 1971a,b). Temperate and tropical copepods, which undergo variation in food supply on a smaller, but more frequent, time scale, store a smaller amount of lipids which are primarily in the form of triacylglycerides (Lee and Hirota 1973; Lawrence 1976).

The importance of lipid stores during periods when food availability is low is suggested by the direct relationship between feeding conditions and energy storage observed in marine invertebrates (Benson and Lee 1975; Lee 1975; Håkanson 1984), and the enhanced survival of cladocerans with lipid stores during periods of low food supply (Tessier *et al.* 1983; Holm and Shapiro 1984; Cowgill *et al.* 1985b). Lipid stores may also be of importance when the food regime remains fairly constant but energetic needs fluctuate. For example, during reproduction the increased metabolic costs experienced by females may be met by utilizing stored energy reserves which are accumulated during periods of lower metabolic needs. Lipid stores of females are depleted during periods of egg production by cladocerans (Goulden *et al.* 1982; Tessier and Goulden 1982; Tessier *et al.* 1983; Cowgill *et al.* 1984; Holm and Shapiro 1984) and marine copepods (Conover 1962; Benson *et al.* 1972; Lee *et al.* 1972) while reserves of males remain unchanged.

Lipid reserves can also change in response to non-food related stress. Lipid stores of the copepod *Epischura* from two lakes with similar food levels were lower in the lake with elevated heavy metal contamination (Arts and Sprules 1987). The actual mechanism by which lipid stores were reduced was not tested, but may have included inhibited feeding or increased metabolism in the presence of heavy metal contaminants. Predation may also act to reduce lipid-mediated starvation resistance. Arts and Sprules (1988) noted that predatory fish, in selecting for large cladocerans, indirectly selected for smaller absolute lipid stores (because lipid stores were a direct function of body size). These smaller cladocerans in the lakes with fish were therefore more sensitive to starvation during food shortages than their larger counterparts in lakes without fish.

Not only are lipid stores affected by food abundance, but also by its nutritional composition. Fat stores in rats (Lawrence 1976) as well as cladocerans (Cowgill *et al.* 1984) are directly correlated with the amount of dietary fat; as well, lipid class composition of rotifers (*Brachionus*) was found to closely resemble that of the algal food (Ben-Amotz *et al.* 1987).

Intraspecific variation in food quality may result in changes in total lipid content as well as lipid class composition of the zooplankton. Phytoplankton reared under nitrogen-limited (Fogg 1959; Ben-Amotz *et al.* 1985; Parrish and Wangersky 1987) or temperature-limited (Smith and Morris 1980) growth conditions tend to accumulate lipids as the end products of photosynthesis. Because copepods derive triacylglycerides directly from phytoplankton lipids (Sargent and Falk-Petersen 1988), high phytoplankton triacylglyceride levels, such as induced by periods of nutrient limitation, may result in high triacylglyceride content in zooplankton lipid stores.

Triacylglycerides comprise the major portion (>60%) of the lipid content of most freshwater invertebrates (except see Cavaletto *et al.* 1989). In marine copepods, whether lipids are stored primarily as triacylglycerides or as wax esters may reflect feeding and metabolic adaptations to available food (Ohman 1988). Triacylglycerides are more easily mobilized than other storage lipids as a consequence of differences in lipase activity. While wax lipase is activated only under starvation stress, triacylglyceride lipase normally remains active at all times (Benson and Lee 1975). Thus, triacylglycerides are readily available to supplement food energy as food supply fluctuates and, during periods of low food abundance, tend to be utilized first, followed by a slower, delayed utilization of wax esters (Lee *et al.* 1972; Benson *et al.* 1972). Because triacylglyceride abundance in copepods reflects food concentration (i.e., it is quickly depleted during periods of food stress) as well as algal chemistry, triacylglyceride content can be a sensitive indicator of the ability of the available food to meet immediate energetic needs (Clarke *et al.* 1985; Rezeg and James 1987; Sargent and Falk-Petersen 1988).

Thus, both the quantity as well as the composition of lipid stores in copepods can be indicative of food conditions. If the changes in the copepod populations reported in Chapter Two are due to nutritional differences in food supply among the enclosures, an associated variation in lipid stores would be predicted. This study investigates the effects of

interspecific and intraspecific variation in food supply on total lipid content and lipid class composition of *Diaptomus kenai* and *D. leptopus*. Effects of interspecific variation in food on lipid stores are investigated with *Diaptomus* populations from the enclosures described in Chapter Two. Effects of intraspecific variation in food on stored lipids in *Diaptomus* are investigated using a single algal species (*Selenastrum minutum*) reared under two different levels of nitrogen limited growth.

METHODS

Field Study

Interspecific Variations in Food Quality: During the period of June 1986 to October 1987, adult *Diaptomus kenai* and *D. leptopus* were collected for lipid analysis during routine sampling of Shirley Lake and the experimental enclosures (as described in Chapter 2). Immediately upon collection, copepods were isolated in dilute artificial pond water (APW: 10% dilution of the formula of Lynch *et al.* 1986) and sorted according to species, sex, and, if female, reproductive condition (Fig. 2). Copepods were rinsed with distilled water and placed on paper towelling to remove excess surface water. Samples of three to five individuals of *D. kenai* or seven to ten individuals of *D. leptopus* were placed in pre-weighed glass micro-culture tubes and each sample identified according to collection date, number of individuals, species, and reproductive status. Three replicates of each species/reproductive status combination were collected, if population density and reproductive status permitted. The samples were held on dry ice for transport to the lab, where they were dried under nitrogen at 50C and weighed. Lee and Hirota (1973) found no evidence of freezing adversely affecting subsequent lipid extraction, based on comparisons of lipids extracted from fresh animals and those taken from animals that had been stored frozen.

Lipid Analysis: Total lipids were extracted and quantified as a percentage of lipid free dry mass (PLFDM) using the micro-gravimetric extraction technique of Gardner *et al.* (1985), as

follows. Samples were homogenized and the lipid extracted in 2:1 chloroform:methanol (vol/vol). After homogenizing, a volume of 0.9% NaCl equivalent to 20% of the sample volume was mixed into the sample (to separate the water-soluble fraction from the lipid fraction). This technique has been used successfully for analyzing small samples by other investigators (Parrish and Ackman 1983a,b and 1985; Rao *et al.* 1985; Cavaletto *et al.* 1989). The primary drawback of this technique is that it is destructive, requiring that the copepods be killed; therefore, it is not possible to monitor individuals over time. While non-destructive techniques have been developed for cladocerans (Tessier and Goulden 1982) and copepods (Klein Breteler and Gonzalez 1988), these are qualitative measures which permit comparison only on a relative scale.

After extraction, a portion of the purified sample was removed for gravimetric analysis with a micropipette, measured for volume, and transferred to weighing cups. The remaining solvent was evaporated in a drying oven at 60C and the lipids remaining weighed to 0.001 mg on a Cahn electrobalance. The percentage lipid was calculated using the following formula:

$$\% \text{ lipid} = [(WR/L)-B](CV/D)(100), \text{ where:}$$

W = weight of lipid (μg) in weighing cup

R = ratio of length (mm) to volume (μL) of micropipette

L = length (mm) of extract delivered to weighing cup in micropipette

B = blank correction (μg lipid per 100 μL of chloroform:methanol without tissue)

C = volume calibration factor (calculated to be 1.07)

V = initial volume of chloroform in solvent (66.7 μL per 100 μL solvent)

D = dry weight of homogenized sample

For lipid class analysis by thin-layer chromatography with flame ionization detection (TLC-FID) (Parrish 1987; Parrish *et al* 1988), approximately 10 μL of purified extract was drawn

into a 50 µL capillary pipette under a nitrogen atmosphere. The volume of the sample was measured and the tube flame-sealed under nitrogen and stored at 0C until TLC-FID analysis. Previous research has indicated that lipid decomposition is not a problem during storage if the samples are stored under nitrogen (Gardner *et al.* 1985; Cavaletto *et al.* 1989).

For TLC-FID analysis, the lipid subsample was spotted directly onto silica-coated Chromarods-SII (Ancal Inc.). The rods were then developed in increasingly polar solvent systems to separate out the lipid classes (Parrish 1987). Between developments in each solvent system, the rods were scanned with an Iatroscan Mark IV (Iatron Labs., Tokyo) connected to a Hewlett-Packard 3392A integrator to identify and measure individual lipid classes. Seven lipid classes were detected and measured by this technique (Table X). A lipid standard containing known amounts of one compound from each of the lipid classes (hydrocarbon, sterol ester, triacylglycerol, free fatty acid, alcohol [aliphatic], sterol [alicyclic alcohol], and phospholipid) was used for calibration. The TLC-FID method does not isolate wax esters from sterol esters. Because sterol esters constitute only a very small proportion of zooplankton lipids (Cavaletto *et al.* 1989, for example, detected no sterol esters in lipid samples from three Lake Michigan calanoid copepod species), I refer to the TLC-FID wax ester-sterol ester peak as "wax esters".

Initial extractions and all lipid class analyses were conducted at the National Oceanographic and Atmospheric Administration's Great Lakes Environmental Research Laboratory in Ann Arbor MI.

Laboratory Study

Intraspecific Variations in Food Quality: In investigations of the effects of food quality on grazer characteristics (e.g., feeding behavior), the desired nutritional differences among the food items can often be confounded by associated changes in some other attribute of the food. This problem is especially true with nutrient-induced variation in food quality, where

TABLE X: The seven lipid classes measured by the Iatroscan, listed in order of detection (from least to most polar). (Adapted from Quigley *et al.* 1989)

a) **HYDROCARBONS**

These are the simplest of the lipid compounds, containing only the elements H and C. Known to be important in the structure of the exoskeleton (Hadley 1981), they also occur internally in small patches of undetermined function (Hadley 1985).

b) **WAX ESTERS**

Wax esters are composed of a fatty acid esterified to an aliphatic alcohol (a hydrocarbon chain with a terminal hydroxyl group). Many aquatic animals, particularly marine copepods, rely upon wax esters for long term energy storage (Chapman 1969; Lee *et al.* 1972; Sargent *et al.* 1977) and buoyancy regulation (Lewis 1970; Sargent and Henderson 1986; Kogeler *et al.* 1987; Hagen 1988).

c) **TRIACYLGLYCERIDES**

Triacylglycerides are formed of three fatty acid chains esterified to glycerol (a trihydroxy alcohol). A chemical energy store in most animals, triasylglycerols tend to be more easily mobilized than wax esters and are used for short term fuel needs in marine copepods (Lee *et al.* 1974; Sargent *et al.* 1977; Hakanson 1984; Falk-Petersen *et al.* 1981).

d) **FATTY ACIDS**

Fatty acids consist of a long hydrocarbon chain with a terminal acidic carboxyl group. In addition to being the functional "building blocks" of the more complex lipids, fatty acids can also occur freely, in small quantities, as a normal component of the overall lipid pool (Chapman 1969).

e) **STEROLS**

Sterols are important components of cell membranes and blood plasma lipoproteins and are widespread in animal tissue (Hadley 1985). Crustaceans are incapable of synthesizing sterols and must rely upon dietary sources (Goad 1976; O'Rourke and Monroe 1976).

f) **ACETONE-MOBILE POLAR LIPIDS (AMPL)**

The AMPL class represents the acetone-extractable part of the polar lipid fraction. Glycolipids, common in chloroplasts and present in trace amounts in animal tissue, are an important component of the AMPL class (Hadley 1985).

g) **PHOSPHOLIPIDS**

As the most polar of the lipid classes, phospholipids consist of two fatty acids and a phosphate group esterified to glycerol. They are important constituents of membranes (Lehninger 1975) and are particularly important in crustaceans as the principal circulating (hemolymph) lipid (Gilbert and O'Connor 1970).

changes in internal cell chemistry may be associated with changes in physical properties such as cell size or shape (e.g. Harrison *et al.* 1977; Admiraal and Werner 1983; Admiraal *et al.* 1986). If the physical features of the cells change under conditions of nutrient limitation, feeding may be affected. Calanoid copepods react to and discriminate between food items based on physical and/or chemical cues (Strickler 1982, 1985; Chow-Fraser and Wong 1985; Lehman and Sandgren 1985; Legier-Visser *et al.* 1986; Butler *et al.* 1989) and their feeding could be affected more by changes in the physical properties of the cells than by the changes in cell chemistry. *Selenastrum minutum* was chosen for the laboratory study because it does not change size under conditions of nutrient limitation (Butler *et al.* 1989) and, therefore, does not mask differences in food quality with differences in physical dimensions.

The alga was cultured in artificial medium (Suttle and Harrison 1988a) with NH_4^+ and PO_4^- concentrations modified to 25 and 16 μM, respectively. These modifications, based on data of Elrifi and Turpin (1985), produced N-limited cells at concentrations required for the study (8-10 X 10^8 cells · l^{-1}). The cultures were maintained in constant light (200 μE · m^{-2} · s^{-1}) at 15C. The cells were cultured (semi-continuous, daily dilutions) at 85% and 23% maximum potential growth rate (1.29 d^{-1} at 15C, Butler *et al.* 1989) to yield high growth rate (HiGR) and low growth rate (LoGR) cells, respectively. Experiments were initiated when cell numbers and relative fluorescence (measured daily prior to dilution) varied less than 10% for a three day period, indicating steady state conditions.

Experiments were conducted in 4 L glass aquaria under constant dark at 15C. Nine aquaria were used for each run and the entire experiment was replicated three times. All nine aquaria were filled with 3.6 L of distilled water. Of these, six received a 400 mL mixture of culture medium and either LoGr or HiGR culture to yield 3 aquaria of each cell type at a final cell concentration of 5 X 10^5 cells·ml^{-1}. Previous research indicated that feeding rate of adult female *Diaptomus kenai* on this alga is maximum at 10^4 cells·ml^{-1} (Butler *et al.* 1989). Three aquaria (the starvation treatment) received 400 mL of algal culture medium without

cells. Thus, each tank was identical with respect to water chemistry (i.e. 10% algal culture medium) but differed in available food.

During August and September 1988, copepods were collected from Shirley Lake and transported to the lab where they were sorted according to species, sex, and size. Only gravid females which could be easily classified as either large or small were used in the study. Because the age and condition of copepods is difficult to assess, the use of gravid females (which, by virtue of being gravid are assumed to be neither senescent nor unhealthy) reduces the possibility of obscuring the results by using copepods of indeterminant age and condition. Once sorted, the copepods were divided into groups of 10 which were then randomly assigned to 3 groups each of 100 large *D. kenai*, 100 small *D. kenai*, and 200 *D. leptopus*, one group for each food treatment. Fifty percent of the medium in each aquaria was replaced daily with fresh algal culture, medium, and distilled water to bring cell numbers to the required density. Copepods were removed from the aquaria at two day intervals in three groups of five (*D. kenai*) or three groups of ten (*D. leptopus*) for total lipid analyses as described above. Each experiment was run until all copepods had been sampled (between 10d and two weeks). This time period would be sufficient to observe a change in lipid stores, because freshwater copepods typically store the majority of their lipids as triacylglycerides (which are readily mobilized).

Statistical Analyses

Total Lipid Content: Total lipid content (percent of lipid-free dry mass, PLFDM) of *D. kenai* collected from the lake and enclosures was analyzed using two-way ANOVA of sex (male, nonreproductive females, and reproductive females) and enclosure with date as a covariate. Changes due to food quality in total lipid content (PLFDM) of large *D. kenai*, small *D. kenai*, and *D. leptopus* in the laboratory were analyzed within each species group by ANOVA of food treatment with time as a covariate. Regression of PLFDM on time analyzed changes in lipid

stores over time in the enclosures and the laboratory. Mean PLFDM (sexes and dates combined) were compared among the enclosures using one way ANOVA with LSD comparisons of the means. PLFDM of *D. leptopus* was analyzed with ANOVA of enclosure effects with date as a covariate. Data from 1986 and 1987 were analyzed separately. All data were log transformed to meet the assumptions of normality and homoscedasticity.

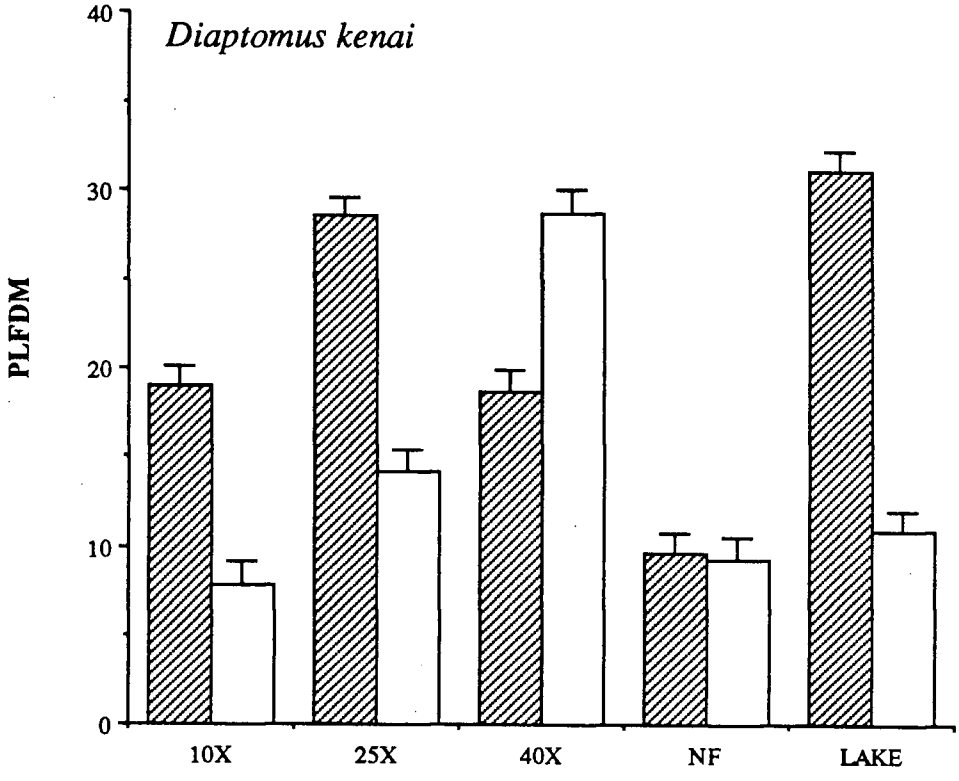
Lipid Class Composition: Lipid class composition data were compared using two-way ANOVA of enclosure and species effects with time as a covariate. Analysis was confined to three lipid groups: wax esters and triacylglycerides (the storage lipids) and polar lipids (the structural lipids). Regression of triacylglyceride composition on time analyzed temporal changes in composition within each enclosure. Lipid class composition data were available only for the 1986 samples. Data were log transformed to meet the assumptions of normality and homoscedasticity.

RESULTS

Field Study

Total Lipids: Figure 14 displays mean lipid content based on lipid-free dry mass for adult *D. kenai* and *D. leptopus* (dates and sexes pooled). ANOVA comparisons of the effects of sex and enclosure on percent lipid-free dry mass (PLFDM) with date as a covariate indicate that there was a significant enclosure effect on PLFDM of *Diaptomus kenai* in 1986, but no significant effect in 1987 (Table XI). There was no significant covariate effect of date within either year, indicating the pattern of change in PLFDM over time was not significantly different among enclosures for either species. Subsequent regression analysis of PLFDM on date indicated no significant time effect in either 1986 ($F = 0.504$, $p = 0.48$ for *D. kenai*; $F = 0.589$, $p = 0.47$ for *D. leptopus*) or 1987 ($F = 2.231$, $p = 0.14$ for *D. kenai*; $F = 0.001$, $p = 0.98$ for *D. leptopus*). One-way ANOVA with LSD indicated that, in 1986, PLFDM of *D. kenai* in the three fertilized enclosures was greater than in the "No Fertilizer" enclosure,

FIGURE 14: Total lipid content, with standard error bars, of adult *Diaptomus kenai* and *D. leptopus* during 1986 and 1987 (lake and "nutrient" enclosures). (shaded columns = 1986; open columns = 1987)


Diaptomus kenai
PLFDM
40
30
20
10
0
10X
25X
40X
NF
LAKE

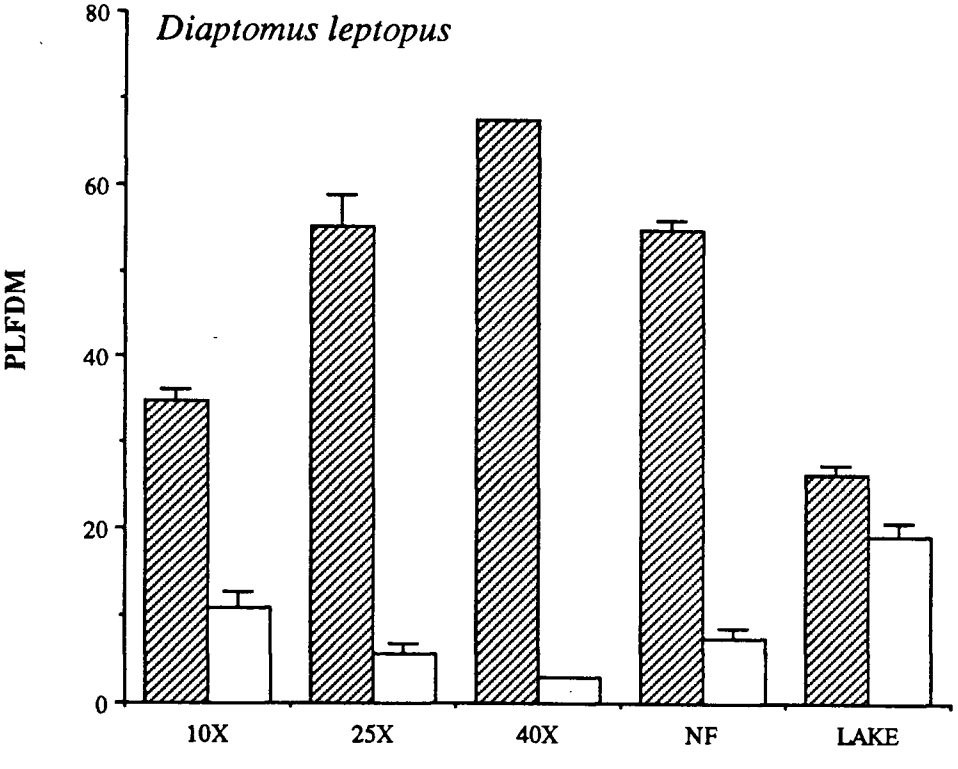

Diaptomus leptopus
PLFDM
80
60
40
20
0
10X
25X
40X
NF
LAKE

TABLE XI: Analysis of *Diaptomus kenai* percent lipid-free dry mass (PLFDM) in the lake and enclosures in a) 1986 and b) 1987, with main (sex and enclosure) and covariate (date) effects.

ANOVA TABLE

a) 1986

Source	Sum Squares	DF	F	P <
Sex	0.589	2	6.916	0.01
Enclosure	0.687	4	4.035	0.01
Interaction	0.180	8	0.022	0.80
Date	0.026	1	0.614	0.50
Residual	3.235	76		

b) 1987

Source	Sum Squares	DF	F	P <
Sex	0.280	2	2.429	0.20
Enclosure	0.533	4	2.310	0.10
Interaction	0.339	8	0.980	0.50
Date	0.155	1	2.679	0.20
Residual	1.154	20		

but less than that observed in the lake (Table XIIa). In 1987, PLFDM of *D. kenai* in the 40X enclosure was significantly greater than in the lake and the other enclosures, which did not differ among themselves (Table XIIb). PLFDM of *D. leptopus* was not significantly affected by any of the factors tested (Table XIII) in either year.

Lipid Class Composition: The abundances of storage and structural lipids (wax esters, triacylglycerides, and polar lipids) in *D. kenai* were compared among the lake and enclosures (Fig. 15a). There were significant enclosure and covariate (date) effects on triacylglyceride (storage) and polar (structural) lipid content of *D. kenai* but not on wax ester content (Table XIV). Linear regression analysis of temporal changes in abundance of wax esters, triacylglycerides, and polar lipids in *D. kenai* (Fig. 16) revealed a significant time effect only for triacylglyceride content in the No Fertilizer enclosure ($R = -0.869$, $P < 0.001$). Lipid class data for *D. leptopus* are presented in Fig. 15b. These data could not be analyzed due to small sample size; however, it would appear that lipid class composition differs considerably among the lake and enclosures.

Laboratory Study

Total lipid content (PLFDM) data from copepods maintained for 14 d on either a starvation, low nitrogen *Selenastrum minutum*, or high nitrogen *S. minutum* diet are presented in Fig. 17. ANOVA comparisons of total lipid content (expressed as PLFDM) indicated significant differences among the three groups of copepods tested (small *D. kenai*, large *D. kenai*, and *D. leptopus*) but no significant treatment effect and no significant interactions between species and treatment (Table XV). A significant effect of the time covariate indicated differences among the treatment and species groups in the way PLFDM changed over time. Subsequent regression analysis indicated that the observed changes, while significantly different when compared among the groups, were significant only within the large *D. kenai* feeding on the low nitrogen food (Table XVI).

TABLE XII: One-way analysis of *Diaptomus kenai* PLFDM in the lake and enclosures in a)1986 and b) 1987, with LSD test for significant differences (* indicates pairs different at the 0.05 level).

a) 1986

ONE-WAY TABLE

Source	DF	SS	F	P <
Between Groups	4	1.383	7.893	0.001
Within Groups	43	1.883		

LSD TABLE

Mean	Group	NF	40X	10X	25X	LAKE
0.983	NF					
1.271	40X	*				
1.276	10X	*				
1.456	25X	*				
1.491	LAKE	*	*	*		

b) 1987

ONE-WAY TABLE

Source	DF	SS	F	P <
Between Groups	4	0.685	1.757	0.20
Within Groups	38	3.703		

LSD TABLE

Mean	Group	NF	40X	10X	25X	LAKE
0.896	10X					
0.966	NF					
1.037	LAKE					
1.150	25X					
1.458	40X	*				

TABLE XIII: Analysis of *Diaptomus leptopus* percent lipid-free dry mass (PLFDM) in the lake and enclosures in a) 1986 and b) 1987, with main (sex and enclosure) and covariate (date) effects.

ANOVA TABLE

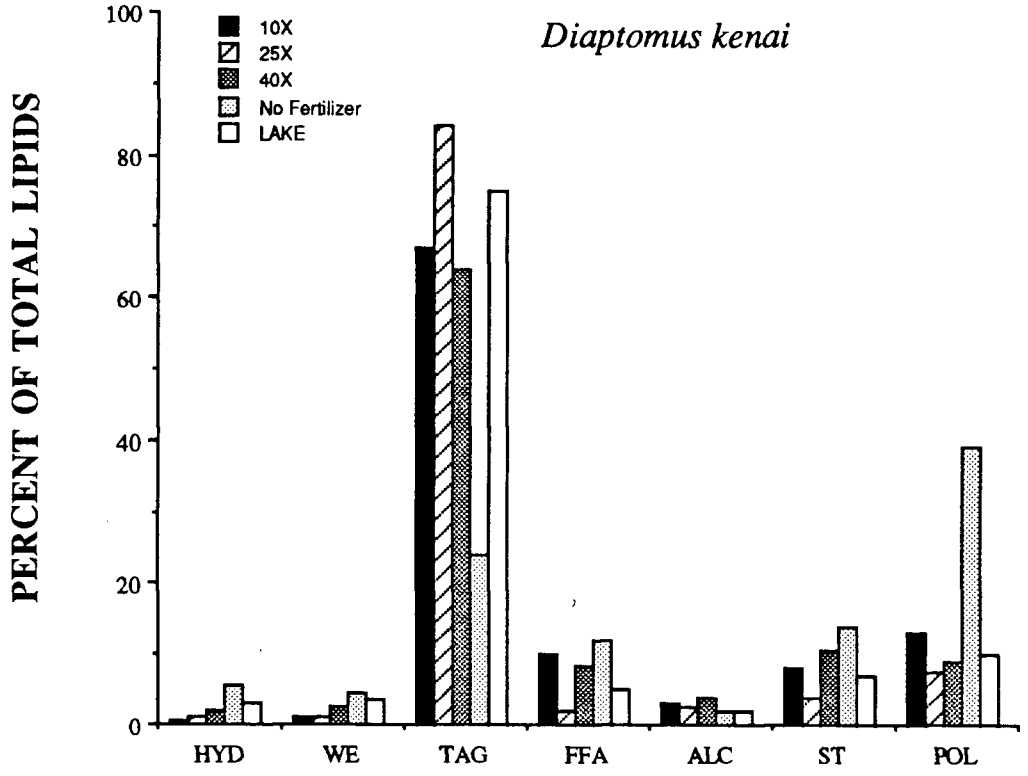
a) 1986

Source	Sum Squares	DF	F	P <	R
Model	0.240	5	0.264	0.90	0.25
Enclosure	0.174	4	0.239	0.90	
Date	0.066	1	0.365	0.60	
Residual	0.728	4			

b) 1987

Source	Sum Squares	DF	F	P <	R
Model	1.006	5	0.757	0.60	0.28
Enclosure	1.005	4	0.946	0.50	
Date	0.000	1	0.001	0.90	
Residual	2.657	10			

FIGURE 15: Lipid class composition of adult *Diaptomus kenai* during 1986 (lake and "nutrient" enclosures). (HYD: Hydrocarbons; WE: Wax Exters; TAG: Triacylglycerides; FFA: Free Fatty Acids; ALC: Alcohols; POL: Polar Lipids)


Diaptomus kenai
PERCENT OF TOTAL LIPIDS
10X
25X
40X
No Fertilizer
LAKE
100
80
60
40
20
0
HYD
WE
TAG
FFA
ALC
ST
POL

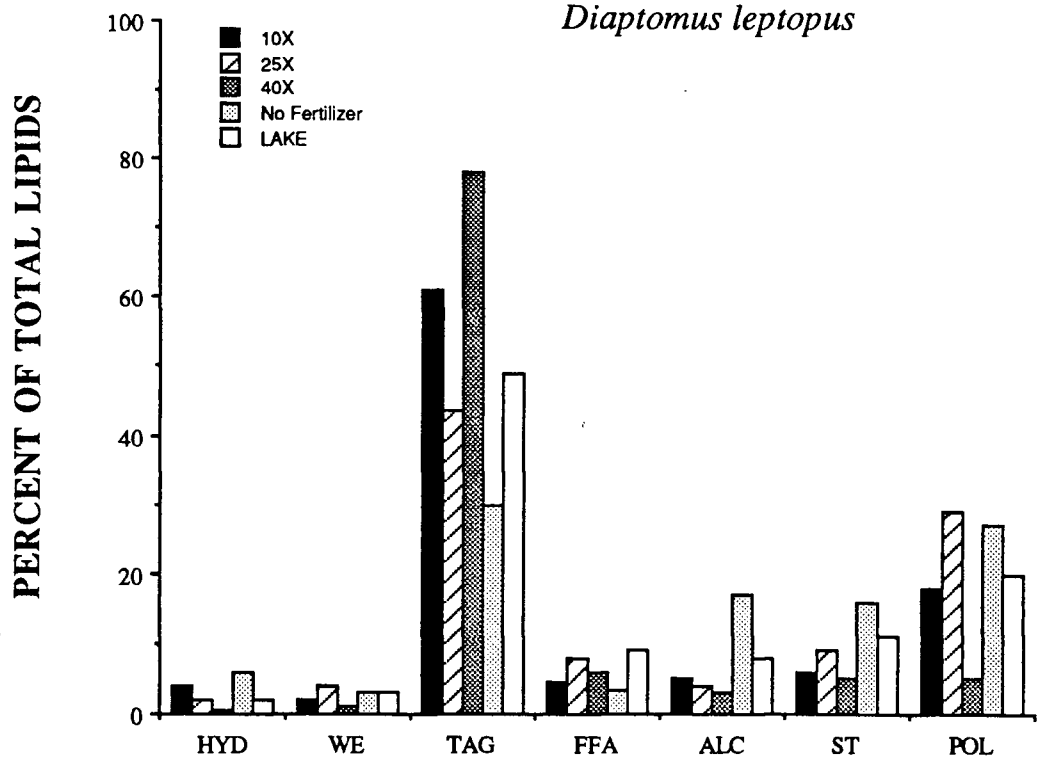

Diaptomus leptopus
PERCENT OF TOTAL LIPIDS
10X
25X
40X
No Fertilizer
LAKE
100
80
60
40
20
0
HYD
WE
TAG
FFA
ALC
ST
POL

TABLE XIV: Analysis of lipid class composition of *Diaptomus kenai* in the lake and enclosures with main (enclosure) and covariate (date) effects for a) percent wax ester, b) percent triacylglyceride, and c) percent polar lipids.

ANOVA TABLE

a) WAX ESTERS

Source	Sum Squares	DF	F	P <	R
Model	0.562	5	0.466	0.80	0.03
Enclosure	0.369	4	0.383	0.82	
Date	0.193	1	0.800	0.37	
Residual	19.522	81			

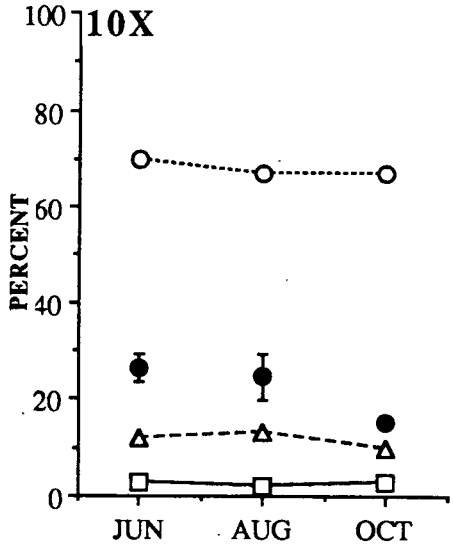
b) TRIACYLGLYCERIDE

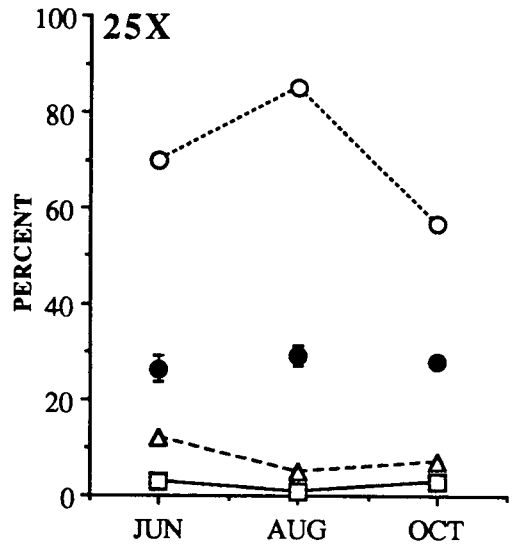
Source	Sum Squares	DF	F	P <	R
Model	1.170	5	5.407	0.001	0.25
Enclosure	0.447	4	2.584	0.05	
Date	0.722	1	16.700	0.001	
Residual	3.504	81			

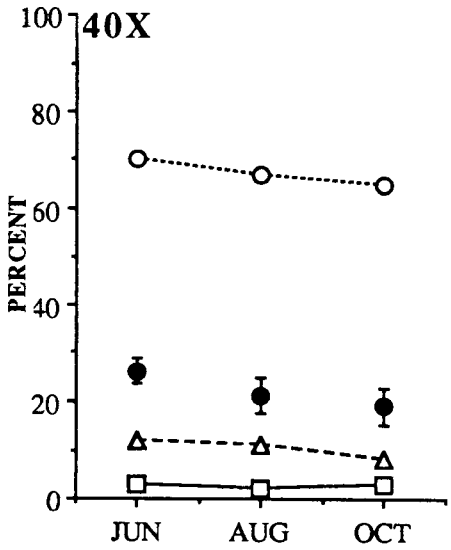
c) POLAR LIPIDS

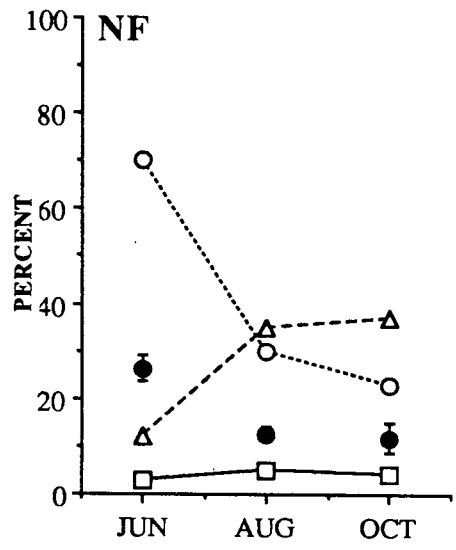
Source	Sum Squares	DF	F	P <	R
Model	1.390	5	2.833	0.05	0.15
Enclosure	0.885	4	2.254	0.07	
Date	0.505	1	5.150	0.05	
Residual	7.949	81			

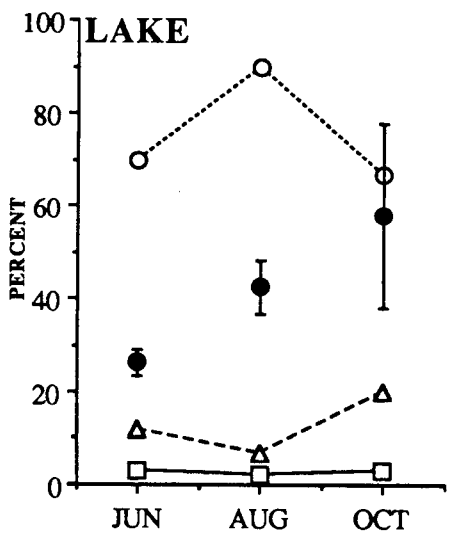
FIGURE 16: Changes in lipid class composition of adult *Diaptomus kenai* during 1986 (lake and "nutrient" enclosures). (= Wax Exters; = Triacylglycerides; = Polar Lipids; = Total Lipids)


10X
100
80
60
40
20
0
PERCENT
JUN
AUG
OCT


25X
100
80
60
40
20
0
PERCENT
JUN
AUG
OCT


40X
100
80
60
40
20
0
PERCENT
JUN
AUG
OCT


NF
100
80
60
40
20
0
PERCENT
JUN
AUG
OCT


LAKE
100
80
60
40
20
0
PERCENT
JUN
AUG
OCT

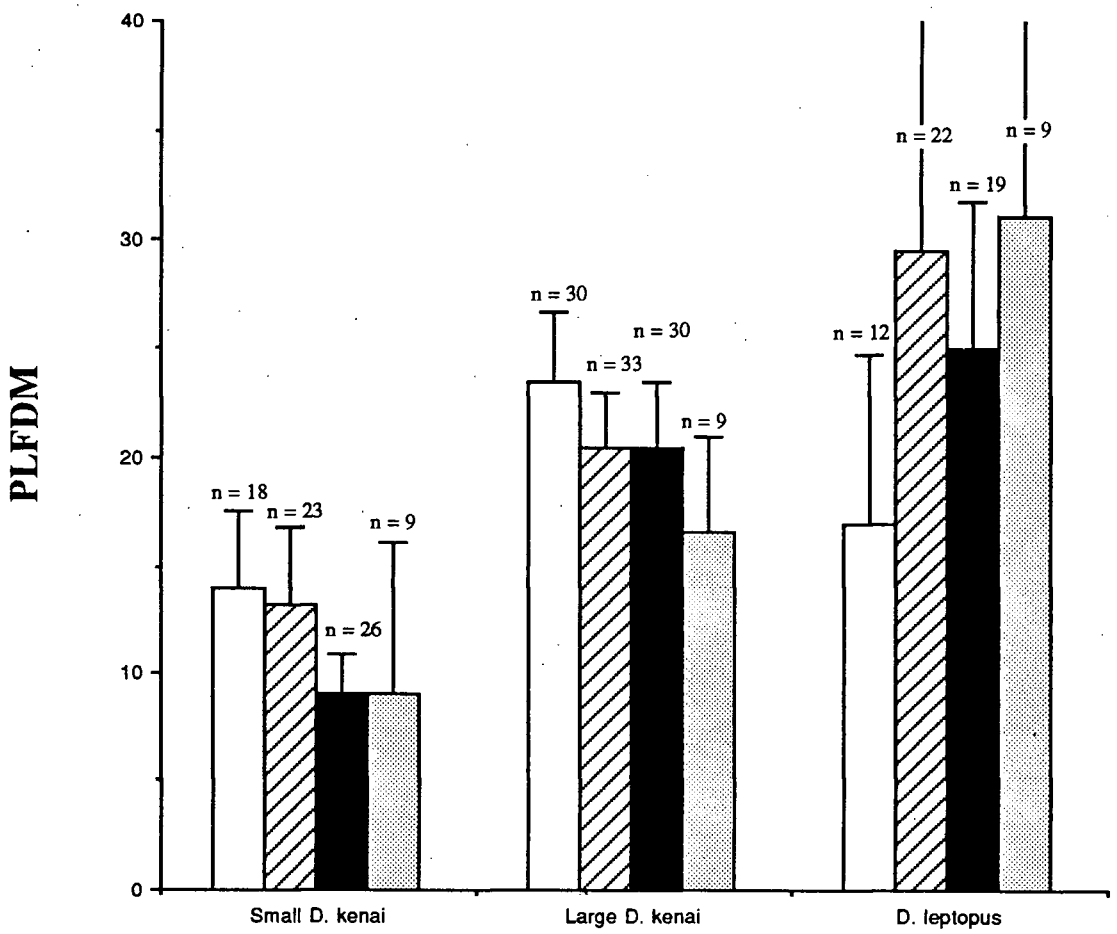


FIGURE 17: Lipid content (as percent lipid-free dry mass, or PLFDM) of adult *Diaptomus kenai* and *D. leptopus* maintained in the laboratory on no food (open columns), *Selenastrum minutum* cultured at low dilution rates (hatched columns), and *S. minutum* cultured at high dilution rates (filled columns), with standard error bars. Speckled bars indicate initial lipid content.

TABLE XV: Analysis of PLFDM of large and small *Diaptomus kenai* and *D. leptopus* in the laboratory feeding study with main (copepod and food type) and covariate (time) effects.

ANOVA TABLE

Source	Sum Squares	DF	F	P <	R
Model	8.269	9	3.442	0.001	0.12
Copepod	6.356	2	11.906	0.001	
Food	0.213	2	0.399	0.70	
Interaction	0.710	4	0.665	0.60	
Time	1.004	1	3.761	0.05	
Residual	54.188	203			

TABLE XVI: Regression analysis of PLFDM versus time for copepods feeding on each of the food types. (Low N: *Selenastrum minutum* cultured at low dilution rate; High N: *S. minutum* cultured at high dilution rate)

a) Small *Diaptomus kenai*

	R	F	P <	N
No Food	0.324	1.874	0.20	18
Low N	0.087	0.160	0.70	23
High N	0.118	0.340	0.60	26

b) Large *Diaptomus kenai*

	R	F	P <	N
No Food	0.331	3.446	0.07	30
Low N	0.359	4.596	0.05	33
High N	0.326	3.334	0.08	30

c) *Diaptomus leptopus*

	R	F	P <	N
No Food	-0.415	2.084	0.20	12
Low N	-0.086	0.148	0.70	22
High N	0.032	0.018	0.90	19

DISCUSSION

The observed variation in total lipid content (PLFDM) among *D. kenai* from the lake and enclosures suggests that there may indeed be variation in the food regime. While total lipid content is often a reflection of food concentration (e.g. copepods: Ikeda 1974; Lee 1974a; prawns: Whyte *et al.* 1986; and oyster spat: Gallager and Mann 1986), variation in *D. kenai* lipid content in 1986 was independent of total cell concentration in the enclosures. *D. kenai* in the three enclosures which were fertilized (which ranged in maximum cell density from 2 to 10 X 10^3 cells·ml^{-1}) accumulated greater lipid stores than those in the nonfertilized enclosure (with a maximum cell density of 4 X 10^3 cells·ml^{-1}), but less than those in the lake (maximum cell density 200 cells·ml^{-1}). The lack of association with cell density is further supported by the fact that while cell density in the enclosures varied significantly with time, total lipid content of copepods in the enclosures was independent of time. Therefore, it would appear that lipid stores in *D. kenai* may be more a function of differences among the lake and enclosures in food quality rather than food quantity. The lack of a significant effect in 1987, however, suggests that there was no longer a substantial difference in food quality among the enclosures, despite the fact that fertilization continued through 1987. The 1987 *D. kenai* populations had very different size-class structures among the enclosures (see Chapter Two). It is possible that the different size classes of *D. kenai* (present in all the enclosures in 1986) have different food requirements, based upon such criteria as chemical composition or ingestibility, and this difference in their ability to utilize the food regime was reflected in their reproductive output (as suggested by the enclosure effect on clutch size presented in Chapter Two). Thus, the population found in the enclosures in 1987 presumably represented the offspring of those copepods which were most able to utilize the 1986 phytoplankton community.

There was also considerable variation in *D. leptopus* lipid content among the enclosures in

1986, suggesting that differences in food supply affected their lipid stores. However, PLFDM was also extremely variable among replicates within enclosures; as a consequence, there was no statistically supported evidence of an enclosure effect on PLFDM for *D. leptopus*.

As with the majority of freshwater copepods, both *D. kenai* and *D. leptopus* in Shirley Lake primarily store their lipids (25 to 85% of total stores) as triacylglycerides. For *D. leptopus*, there was no statistically significant variation in lipid class composition among the enclosures and lake, although the data suggest variation in realized food quality among the enclosures. For *D. kenai*, however, there was a significant enclosure effect on lipid class composition, particularly in triacylglyceride content. Triacylglyceride content of *D. kenai* in the lake varied considerably over time, with maximum levels (nearly 90% of total lipid stores) occurring in August. This peak most likely reflects decreased energy demands (it coincides with a period when small *D. kenai* are ending reproduction, but large *D. kenai* have not yet initiated reproduction) as well as natural changes in nutrient levels and phytoplankton community composition, rather than changes in absolute cell density. In the fertilized enclosures, which recieved a constant supply of nutrients, variation in the food quality could probably be attributed to differences in species composition. In the 10X and 40X enclosures, triacylglyceride content of the copepods was fairly constant over time (varying less than 5%) while triacylglyceride content of copepods in the 25X enclosure varied in a pattern similar to that in the lake. However, in the unfertilized enclosure, which did not receive a steady supply of nutrients, *D. kenai* triacylglyceride content decreased steadily over the course of the summer.

Results of the laboratory feeding study suggest that, while there were differences among the three copepod groups (small *D. kenai*, large *D. kenai*, and *D. leptopus*) in total lipid content, there was no variation in lipid content attributable to differences in *Selenastrum minutum* due to nitrogen limitation. The only discernable effect of diet was the decrease in lipid stores of *D. leptopus* under starvation conditions; however, variations in algal nutrient limitation did not

affect lipid content in any of the tested copepods. The fact that *D. kenai* is capable of discriminating between and selectively ingesting *S. minutum* cells according to degree of nitrogen limitation (Butler *et al.* 1989) suggests that there are chemical differences between the two cell types, but these differences do not appear to be reflected in lipid content. Although lipid content of the two algal cultures was not measured, algae raised under nitrogen limited conditions are known to have a high lipid content (Reynolds 1984; Parrish and Wangersky 1987). However, *D. leptopus* and *D. kenai* stores do not seem to reflect this intraspecific variation in food chemistry. It is possible that, because the food levels maintained during the experiment were in excess of satiation food density, the copepods offset food quality affects by altering ingestion rates, thereby maintaining lipid stores.

If lipid content cannot be attributed to overall food density or intraspecific variation in cell chemistry (induced by different levels of nutrient limitation), then lipid content possibly reflects differences among the lake and enclosures in phytoplankton species composition and in the ability of the copepods to utilize the phytoplankton community. The next chapter investigates feeding rates and selectivities associated with variations in phytoplankton density and composition.

Chapter Four

FOOD SELECTION BY DIAPTOMUS KENAI: EFFECTS OF FOOD COMPOSITION AND DENSITY

In the previous chapter, I addressed the effects of food on overall nutritional status, particularly with respect to energy stores. I discussed food effects in terms of food abundance and in terms of food quality (e.g., cell chemistry). However, food quality is not only a function of the chemistry or energetic content of the cell, but is also a function of the accessibility of that food energy, including energetic costs associated with feeding. For example, if the energy spent acquiring a food item exceeds the energy gained upon ingesting it, that food item will be of lower quality than one from which the consumer gains more energy ingesting it than was spent capturing it.

This idea of food quality based upon the costs and benefits associated with a food item was conceptualized in a model presented by Charnov and Orians (1973), a modification of Holling's (1966) four components of foraging (based on time allocation). Their model takes into account a food item's energetic value (based on energy costs and gains) and capture probability associated with each of the four feeding subprocesses: search, pursuit, attack, and ingestion, thus summarizing the energetic value of a food item as a combination of its detectability, "catchability", ingestibility, and digestibility.

From this model, one can see that the rate of ingestion of a particular food item is not necessarily constant and will vary according to a wide range of factors. Detectability can be affected by such characteristics of the food as size or smell (which influence the consumer's ability to locate the food) or density (which affects encounter rate). "Catchability" is affected by the size of the food item as well as by its shape; together these affect the consumer's ability to effectively take, retain, and manipulate a particular food item. As with

"catchability", ingestibility is affected by size and shape of the food (which determine the ability of the consumer to maneuver the captured food into its mouth), but smell and/or taste will also influence ingestibility. Once the food has been successfully detected, captured, and ingested, the final step in the feeding process is digestion of the food, which is affected by such factors as the presence or absence of a indigestible covering around the food or the presence of appropriate digestive enzymes.

In the phytoplankton community, nearly all of these factors could be affected by changes in the nutrient regime (quantity and composition). The internal chemical composition of the cell, which may affect its "taste", varies with the degree, and form, of nutrient limitation (e.g., Fogg 1959; Ben-Amotz *et al.* 1985; Dortch 1987; Parrish and Wangersky 1987), as does the external chemistry, or "smell", of a cell (e.g., Admiraal and Werner 1983; Admiraal *et al.* 1986). For a number of phytoplankton species, cell size and shape vary with nutrient levels (e.g., Turpin and Harrison 1980; Suttle *et al.* 1987). Effects of nutrient levels on growth rates and cell densities have been well documented (see review by Kilham and Kilham 1984). Thus, temporal and spatial fluctuations in lake nutrient levels (discussed in Chapter One) could have pronounced effects on attributes of the food regime which affect feeding behavior of zooplankton grazers.

The effect of food concentration on the feeding process has been well studied for a wide variety of organisms. In general, the relationship (or functional response) between ingestion rate and food concentration can be expressed in curvilinear and/or rectilinear models (Holling 1965; Mullin *et al.* 1975). The predictions of these models have been verified in laboratory studies of zooplankton feeding on single foods presented over a range of food densities (O'Connors *et al.* 1980; Porter *et al.* 1982; Stemberger 1986; Dagg and Walser 1987), with satiation occurring at certain concentrations. However, studies of Conover (1978) and Mayzaud and Poulet (1978) failed to demonstrate satiation for five copepod species when offered natural seston over a range of concentrations similar to what would be

observed in nature. Based upon these results, Conover (1978) argued that satiation is a laboratory artifact reflecting sudden exposure to unnaturally high concentrations of a single food source. Buckingham (1978) also did not observe satiation in *Diaptomus* feeding until seston concentrations exceeded natural densities, but noted that conditioning the animals to different food densities prior to conducting the feeding studies did not affect the satiation response. Thus, in her study, satiation could not be viewed as a artifact of "sudden" exposure to abnormally high food densities.

The influence of food size and shape on feeding is intuitively obvious - small food may be harder to locate and capture, and large food, while easily located, may be harder to manipulate and ingest. In fact, the traditional view of copepod feeding centered on the size issue. Copepods were perceived as passive feeders capturing food by filtering water through their maxillary setae, with the size of the holes between the setae restricting the diet according to cell size. Given this view of the feeding process, researchers tended to concentrate more on physical attributes of the food (e.g., cell size and shape, or density) affecting copepod feeding (Frost 1972, 1977; Nival and Nival 1973, 1976, 1979; Boyd 1976). Coupling the view of copepods as passive filter feeders with Holling's model of the feeding process, which assigns a fair amount of importance to cell size/shape, researchers viewed cell size distribution as a measure of food availability in nature.

Although cell morphology and abundance remain arguably important aspects of copepod feeding, high-speed microcinematography (Alcaraz *et al.* 1980; Koehl and Strickler 1981; Strickler 1982; Paffenhöfer *et al.* 1982) has revealed that calanoid copepod feeding is far more directed and less passive than previously thought. Research into copepod feeding mechanisms, particularly with adult *Calanus* and *Diaptomus*, has demonstrated that copepods can capture food particles by any of three different processes, depending upon the size of the food item, and can switch among these behaviors quite readily. Small particles (less than 10 μm equivalent spherical diameter: ESD) are captured passively (without detection) through

low amplitude movements of the appendages, trapped by the first and second maxillae, and transferred to the mouth (Cowles and Strickler 1983; Price *et al.* 1983; Vanderploeg and Paffenhöfer 1985; Price and Paffenhöfer 1986). Larger animal prey (such as rotifers around 100 μm ESD) are captured by a lunging motion, involving rapid movements of the antennae that propel the copepod toward its prey (Williamson and Vanderploeg 1988). But copepods detect and react to intermediate sized particles (such as large algal cells ranging between 10 - 100 μm ESD) from a distance (Vanderploeg and Paffenhöfer 1985), and actively capture the cells with directed feeding movements which bring the cells into capture range (Koehl and Strickler 1981). Studies of flow dynamics and chemical "envelopes" around food particles lend support to the theory that copepods react to chemical stimuli in locating and capturing food particles (Andrews 1983). Although mechanoreception is most likely an influencing factor (Legier-Visser *et al.* 1986), the chemical basis of feeding has become a popular research topic in laboratory zooplankton studies, with researchers demonstrating that feeding can be highly sensitive to the chemical attributes of food particles. Thus the feeding process becomes more complex, for not only must one consider the physical attributes of the cells, but the chemical attributes as well.

The biochemical attributes of a cell can influence food selection in two ways, affecting the "taste" and/or the "smell" of the food. Under conditions of high nutrient levels (i.e., when cells are not growing under nutrient limited conditions), cells tend to "leak" a greater quantity of extra-cellular metabolites (Admiraal and Werner 1983; Admiraal *et al.* 1986). As a consequence, there is a larger chemical envelope around a cell, increasing the likelihood of detection. Moreover, the internal chemistry of the cell (e.g., lipid pools and protein levels) can change with nutrient limitation (Dortch 1987; Furnadzicva *et al.* 1987; Houde and Roman 1987; Parrish and Wangersky 1987). If internal chemistry is affected, then "taste" and subsequent ingestion of the cell will also be affected. Thus, chemically mediated feeding can potentially involve two processes - long range detection and short range "taste".

We now recognize the potential for calanoid copepods to be highly discriminating when feeding, with ingestion influenced by such criteria as nutritional value of a food item or chemical characteristics of a particle. Copepods can discriminate between, and selectively ingest, foods based upon interspecific differences. For example, copepods have higher selectivities for high quality or non toxic algal species than for lower quality or toxic species (Fulton 1988; DeMott 1989; Vanderploeg *et al.* 1990). Similarly, *Acartia*, offered a mix of *Skeletonema* and *Phaeocystis*, selectively ingested *Skeletonema*, which was demonstrated to be the higher quality food in terms of reproductive output (Verity and Smayda 1989). Calanoids are also capable of discriminating between particles based on difference in cell chemistry. Cells grown under nitrogen deficient conditions are ingested at a lower rate than nitrogen sufficient cells by *Acartia tonsa* (Houde and Roman 1987; Cowles *et al.* 1988) and *Diaptomus kenai* (Butler *et al.* 1989). *Eudiaptomus* and *Pseudocalanus* are both highly selective for live algae versus heat killed cells (DeMott 1988). The chemical basis of food selection in copepods is further supported by the fact that they can be "fooled" into ingesting inorganic particles which would ordinarily be ignored. For example, *Diaptomus* (DeMott 1986), *Eudiaptomus* (DeMott 1988), and *Centropages* (Van Alstyne 1986) ingest microcapsules "flavored" with algal exudates more readily than they do unflavored ones.

Given the known variation in abundance and chemical composition of natural algal communities, food consumption by copepods in natural phytoplankton assemblages will be influenced by numerous factors, including variation in inter- and intraspecific food quality, relative and absolute abundance of the various food items, and the number of alternative food choices. Chapter 2 presented information on population-level responses to variation in food composition and quality. The phytoplankton assemblages studied supported copepod populations which differed in such attributes as population size, reproduction, and body size. Chapter 3 investigated patterns of lipid storage in response to changes in food. I concluded that lipid stores in *Diaptomus kenai* and *D. leptopus* were affected by food-species composition and, while the two copepod species differed in their sensitivity to differences in cell

chemistry between cells, there was little difference between the two size classes of *D. kenai* in their response to intraspecific variations in food. This chapter investigates behavioral responses to food composition, and the flexibility of foraging decisions based on food composition and abundance. The ultimate objective is to elucidate behavioral feeding mechanisms of the two size classes of *D. kenai* which could influence their ability to coexist.

METHODS

Since the purpose of this study was to investigate the behavioral flexibility of foraging based on food composition and quantity, a diversity of food regimes was obtained by fertilizing the enclosures (described in Chapter 2) with a range of nitrogen:phosphorus ratios. Beginning in July 1989, the enclosures were fertilized weekly at the levels presented in Table XVII for two months (July - August), after which time phytoplankton were collected for laboratory feeding studies.

Food Composition

A study, using adult female *D. kenai* collected from the lake, of the effects of food composition (i.e., taxonomic composition of the phytoplankton community) on filtering rate and selectivity was conducted with phytoplankton collected from each of the enclosures and the lake. The experiments were conducted over a one-week period during the final week of August, a time when gravid females of both size classes could be collected. Because age and condition of copepods are difficult to assess, use of gravid females (which, by virtue of being reproductive, are assumed to be healthy) ensured that interpretation of the results would not be obscured by using copepods of indeterminate age and condition.

Using the techniques described in Chapter Two, phytoplankton were collected from the lake and enclosures and copepods were collected from the lake. Phytoplankton were collected from a different enclosure every morning over a one-week period until feeding studies had

TABLE XVII: Nutrients added weekly to the enclosures in 1989.

ENCLOSURE	NH_4Cl	KH_2PO_4
10X	1170 mg	300 mg
25X	2920 mg	300 mg
40X	4660 mg	300 mg

been conducted on phytoplankton communities from each enclosure and the lake. Phytoplankton and copepods were kept in separate 4 L plastic buckets at 10 - 15C during transport to the laboratory.

A number of techniques is used to study feeding behavior in zooplankton. These techniques have been reviewed and summarized by Peters (1984). The most commonly used of these are based on one of three techniques: radioactively label the entire food supply and measure rate of label uptake by the grazer; add a single labelled food item to a mixture of foods and measure rate of label uptake by the grazer; and place grazers in a mixture of food particles for a period of time and measure the change in cell concentration. While the first technique provides an accurate assesment of cell uptake in unialgal foods, differences between algal species in label uptake efficiency hinders accurate estimates of actual ingestion. The second technique assumes that ingestion of the labelled cell indicates of ingestion rate for all other components of the food assemblage, which is a potentially erroneous assumption given what is known about copepod discriminatory feeding. The last technique is prone to the errors associated with enclosing grazers in small volumes of water, problems which are accentuated by long incubation times (e.g., breakup of large colonial forms into individual cells, fertilization effects from zooplankton excretion, disturbance effects of containment on copepod feeding behavior). Recognizing the problems inherent with each technique, I elected to use the third method because the labeling techniques would fail to give adequate, and appropriate, information on specific ingestion rates for individual taxa in each phytoplankton community.

The phytoplankton sample was split among nine 250 mL bottles, with a sample preserved in Lugol's to provide information on starting composition. Adult female *D. kenai* which could easily be classified as either large or small were isolated from the copepod samples and sorted into two groups according to body size. The copepods were randomly allocated to the bottles according to size, yielding three bottles each with eight large *D. kenai*, three bottles

each with eight small *D. kenai*, and three bottles with phytoplankton only (controls). The bottles were placed on a plankton wheel rotating at 1 rpm at 15C in total darkness. After six h, the bottles were removed from the wheel, the contents poured through a 200 μm mesh net to remove the copepods, and the remaining phytoplankton preserved in Lugol's solution. Phytoplankton samples were then settled, and taxa identified and counted (as described in Chapter Two). In addition, greatest axial linear dimension (GALD) was measured on 20 cells of each taxon using an ocular micrometer (cell volume estimated from linear dimensions). This procedure was repeated daily until grazing studies had been conducted on phytoplankton communities from the lake and each of the three enclosures.

Food Density

A study of the effects of food density on filtering rate and particle selectivity was conducted with phytoplankton collected from the 40X enclosure and copepods collected from the lake. Samples were collected and transported to the lab as described above. The test conducted the previous day with undiluted phytoplankton from the 40X enclosure (as part of the food composition study described above) served as the full strength test. (Comparisons of the phytoplankton communities collected from the enclosure indicated no change in taxonomic composition between the two days.) Two sequential dilutions (using artificial pond water as described in Chapter Two) of the phytoplankton sample yielded a one-half dilution and a one-quarter dilution of the 40X enclosure phytoplankton community. These dilutions were split among eighteen 250 mL bottles to yield nine bottles of each dilution. Eight large adult female *D. kenai* were added to three bottles of each of the dilutions and eight small adult female *D. kenai* added to three other bottles. The remaining three bottles of each dilution, with only phytoplankton, were controls. The bottles were placed on the plankton wheel, as described above. There was no mortality of the copepods during the course of the study.

Similarity Indices

The Renkonen Index, or Percent Similarity Index, was used to compare the taxonomic composition of the four phytoplankton communities (taken from the lake and the three enclosures), and to compare the diet composition of the large and small *D. kenai* feeding on each of the four phytoplankton communities. This percentage index is based upon the relative abundances of items in two samples or diets and is considered to be one of the best indices for quantitative assessment of similarity or diet overlap (Krebs 1989). It ranges in value from 0 (no similarity) to 100 (complete similarity). The index is calculated as:

$$\mathbf{P = \Sigma\ minimum\ (p_{1i}, p_{2i})}, \text{ where:}$$

P = % similarity between samples 1 and 2

p_{1i} = percentage of species i in sample 1

p_{2i} = percentage of species i in sample 2

Electivity Indices:

Since the introduction of Ivlev's index to measure feeding preferences (Ivlev 1961), researchers have developed various indices to explain and describe foraging and food selection based on modifications of the original index. The most commonly used of these are reviewed by Lechowicz (1982). In this study I used the E^* index of Vanderploeg and Scavia (1979) to measure electivity. This index was recommended by Lechowicz (1982) as being the most useful for comparing feeding preferences among spatially and/or temporally separated sites. As with the original Ivlev index, E^* measures electivity for a food item over a numerical range of -1 (for most avoided) to +1 (for most preferred), with 0 indicating no preference, and takes into account not only the relative abundance of that food, but also the number of alternate food choices. This last characteristic is of particular importance for the purpose of this study because variations in food supply in nature are generally associated with variations in the number of different food items.

E^* was used to measure electivity for each food item under each of the feeding conditions and was calculated by the following algorithm:

$$\mathbf{E^* = [W_i - (1/n)] / [W_i + (1/n)]}, \text{ where:}$$

n = number of kinds of food items

$W_i = [r_i/p_i] / [\Sigma(r_i/p_i)]$

r_i = proportion of food *i* in the diet

p_i = proportion of food *i* in environment

Filtering Rate:

Feeding behavior can also be assessed through filtering rates (clearance rates). As with electivity indices, filtering rate represents a behavioral response to food characteristics such as nutritional content as well as non-nutritional factors (detectability, "catchability", handling ease). Unlike electivity indices, however, filtering rates reflect actual foraging effort and ingestion. For copepods, foraging effort is represented as the amount of water that must be "processed" to encounter the cells ingested. If the amount of water processed is the same for all cell types, then the feeding process can be viewed as non-discriminant, with all cells removed as they are encountered in the water column in equal proportions relative to their abundance. If, however, cells are removed disproportionately, more readily ingested cells will be removed at a greater rate than less readily ingested cells.

Filtering rate was calculated for each algal taxon according to the following equation:

$$\mathbf{FR = ([\ln(C) - \ln(E)]V) / (NT)}, \text{ where:}$$

FR = filtering rate ($ml \cdot copepod^{-1} \cdot d^{-1}$)

C = control cell concentration ($cells \cdot mL^{-1}$)

E = experimental cell concentration ($cells \cdot mL^{-1}$)

V = volume (mL)

N = number of copepods

T = duration of experiment (d)

This formula, modified after the equation of Landry (1981), is applicable to the present study because changes in phytoplankton abundance were attributable to the presence of grazers (i.e., there was no change in abundance in the control jars during the course of the experiments). Preserved phytoplankton samples from the grazing studies were settled for 36 h in settling chambers prior to identification and enumeration on an inverted microscope at 300X. Comparison of starting and final compositions of the controls indicated little or no change during the six hour study period; therefore, electivity indices and filtering rates were calculated comparing cell densities in the controls and treatments at the conclusion of the experiment.

Statistical Analyses

The influence of the various characteristics of the food were analyzed on two levels. The attributes of each individual food taxon affecting the filtering rates and selectivities of the grazer were analyzed first. FR and E^* of large and small *D. kenai* were analyzed in a one-way ANOVA with taxon as the independent variable for individual phytoplankton taxon effects. To assess the influence of physical attributes of the food on feeding, FR and E^* values for large and small *D. kenai* feeding on lake phytoplankton were compared against the independent variables GALD and cell volume with multiple regression. In analyzing both the food density and the food composition studies, multiple regression identified linear associations between the independent variables (defined below) and FR and E^* values for large and small *D. kenai* feeding on each taxon in the phytoplankton communities. In the food density experiment, the independent variables examined were dilution, total cell density of the phytoplankton community, and density of each individual taxon at the start of the experiment, referred to as dilution, total, and density, respectively. The dilution variable provides a measure of variation due to factors associated with dilution beyond the obvious changes in cell concentration (e.g., changes in cell chemistry due to dilution of nutrients in the water). Independent variables examined in the food composition experiment were individual taxon density, total cell density, percent of the total food regime, number of

alternate food choices (i.e., number of phytoplankton taxa), and enclosure (i.e., source of the food community), referred to as taxon, total, percent, number, and enclosure, respectively. The enclsoure variable was included to provide an estimate of variation due to unmeasured attributes of each food community (e.g., differences in "taste" or detectability due to degree of nutrient limitation).

Comparisons of composition of, and feeding on, the phytoplankton communities as a whole required multivariate statistical techniques. While multivariate techniques are used extensively as an analytical tool in terrestrial plant ecology (see Digby and Kempton 1987), they are seldom used in aquatic studies, despite the similar problems faced by researchers (Merrick and Ganf 1988). My aim was to summarize the data while exposing inherent structure within the data; therefore, the following two techniques were used. Hierarchical classification using the Euclidean distance index (a measure of dissimilarity based on distances between pairs of variables) and UPGMA clustering (based upon unweighted arithmetic averages between pair-groups) identified similarities and partitioned the data into discrete groups, which are plotted in dendrograms according to the degree of dissimilarity. Groups are linked along a dissimilarity scale ranging from 1 (least dissimilar) to 25 (most dissimilar). The relationships between taxonomic composition of the four food groups and between large and small *D. kenai* filtering rates and selectivities were examined using discriminant analysis (i.e., canonical variate analysis). Discriminant analysis emphasizes differences among groups to identify those taxa most important for distinguishing among the communities (Digby and Kempton 1987). Using a forward entry algorithm to minimize Wilks' lambda (see Chapter Two), the variables (genera) most important in separating the groups are identified (Norusis 1988).

RESULTS

Figure 18 presents the taxonomic composition of the lake and enclosure phytoplankton communities collected at the time of the feeding experiments, and the taxonomic composition of the phytoplankton ingested by large and small *D. kenai*. Comparisons of phytoplankton composition using the Renkonen Index (which considers only relative abundance of each taxon) indicated that the lake phytoplankton community was similar in composition to the 25X community (P = 83%), but only slightly similar to the 40X community (P = 20%). All other paired comparisons yielded similarity indices of less than 10%, indicating little similarity in community composition. However, hierarchical classification, which in this case incorporates absolute density as well as relative abundance, revealed that the least dissimilar groups were the 10X and lake phytoplankton communities (Figure 19). Dissimilarity values linking replicates of the food treatments were low, indicating little difference among replicates in composition. The 10X, 40X, and lake phytoplankton communities were linked at a dissimilarity index of 4, indicating low dissimilarity. The 25X phytoplankton community joins the other groups at 25, indicating that this community is the most different in composition from the others.

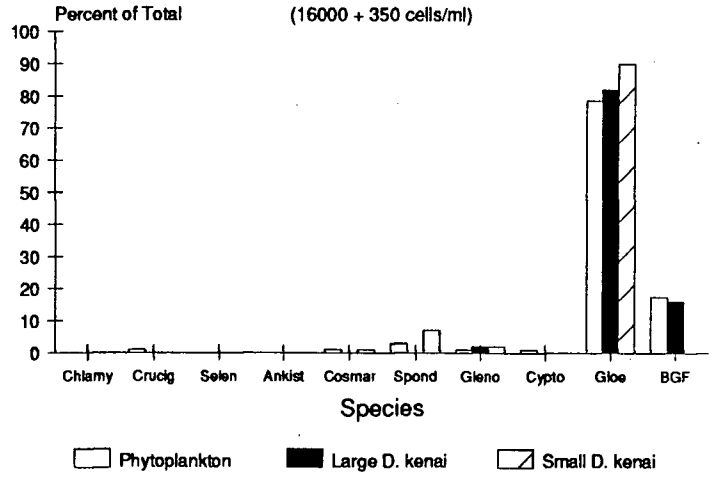
Discriminant analysis separated the four phytoplankton communities on the first two discriminant functions as shown in Figure 20. Nearly 94% of the total variance in composition (Table XVIII) was explained by the first two functions. The forward entry algorithm to minimize Wilks' lambda identified five variables (taxa) as most important in separating the groups: *Ankistrodesmus*, *Selenastrum*, cryptomonads, *Glenodinium*, and *Crucigenia*.

Feeding Behavior and Food Composition

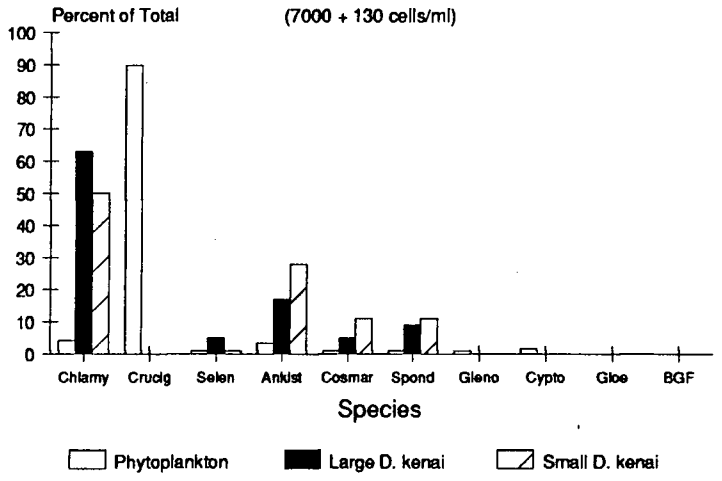
Diet composition of large and small *D. kenai* feeding on phytoplankton from the three enclosures and the lake are presented in Figure 18. Renkonen Index comparisons of the diet

FIGURE 18: Taxonomic composition of lake and enclosure phytoplankton communities (expressed as percent of total community), and of phytoplankton ingested by large and small *D. kenai* (expressed as percent of total diet). (Chlamy: *Chlamydomonas*; Crucig: *Crucigenia*; Selen: *Selenastrum*; Ankist: *Ankistrodesmus*; Cosmar: *Cosmarium*; Spond: *Spondylosium*; Gleno: *Glenodinium*; Crypto: cryptomonads; Gloe: *Gloeotheca*; BGF: filamentous cyanobacteria)

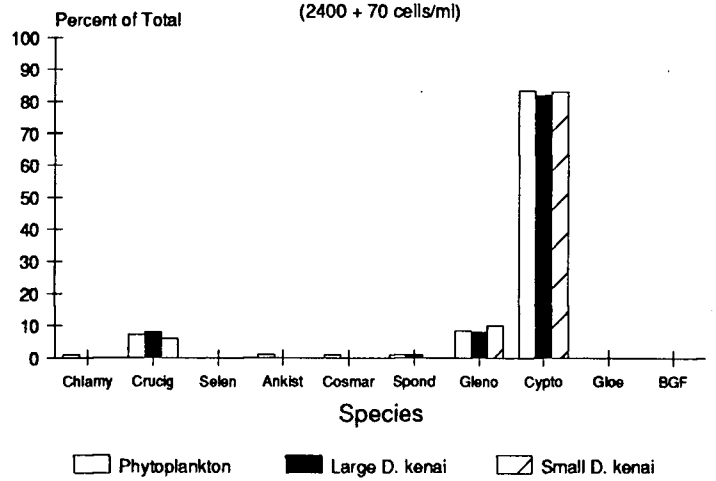
10X Enclosure



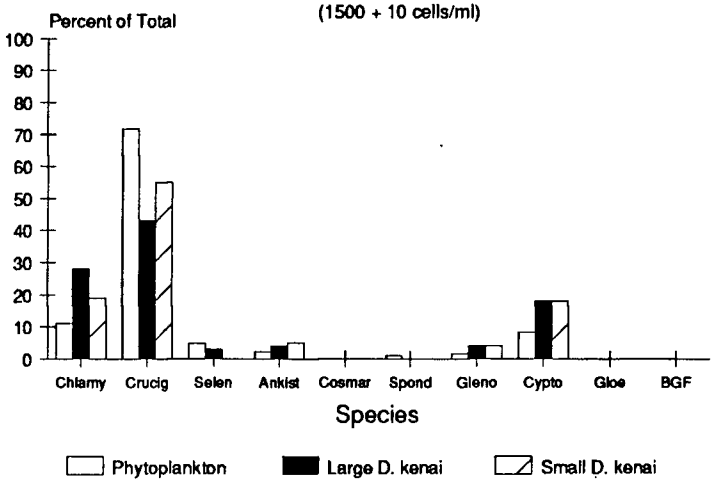
25X Enclosure



40X Enclosure



Lake



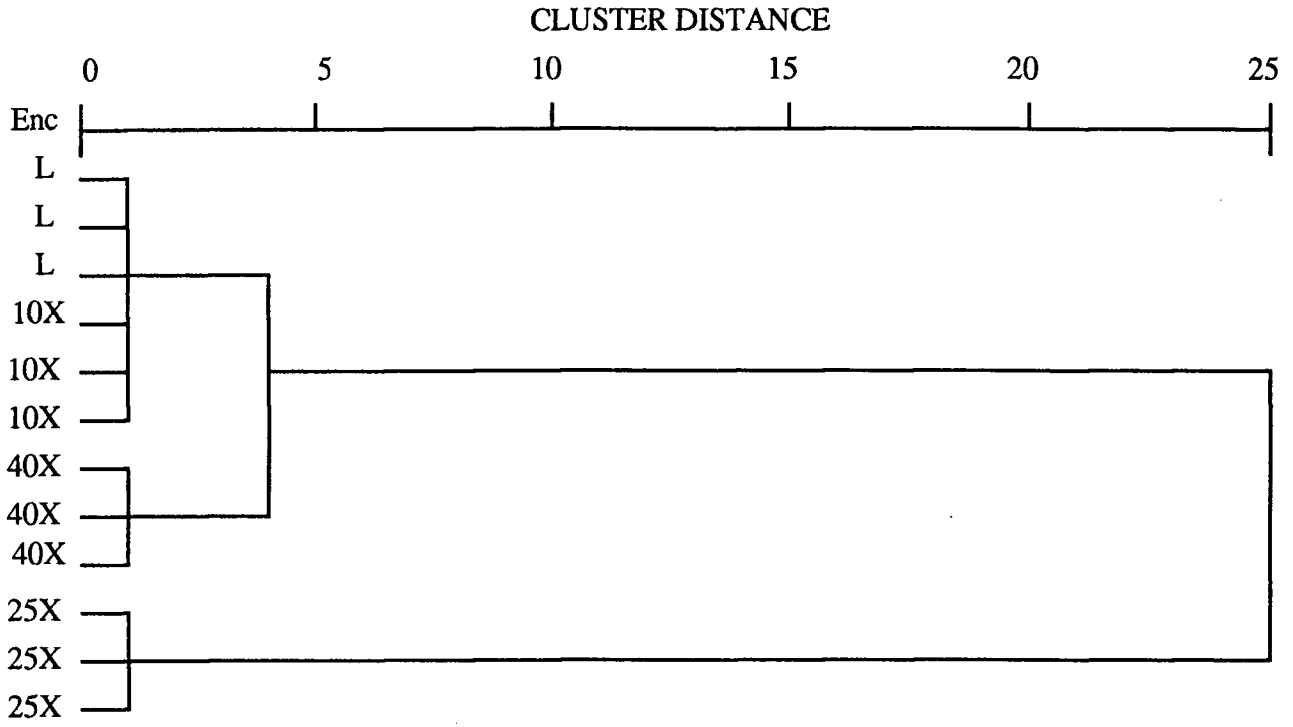


FIGURE 19: Dendrogram of hierarchical classificication of phytoplankton communities in the lake and enclosures.

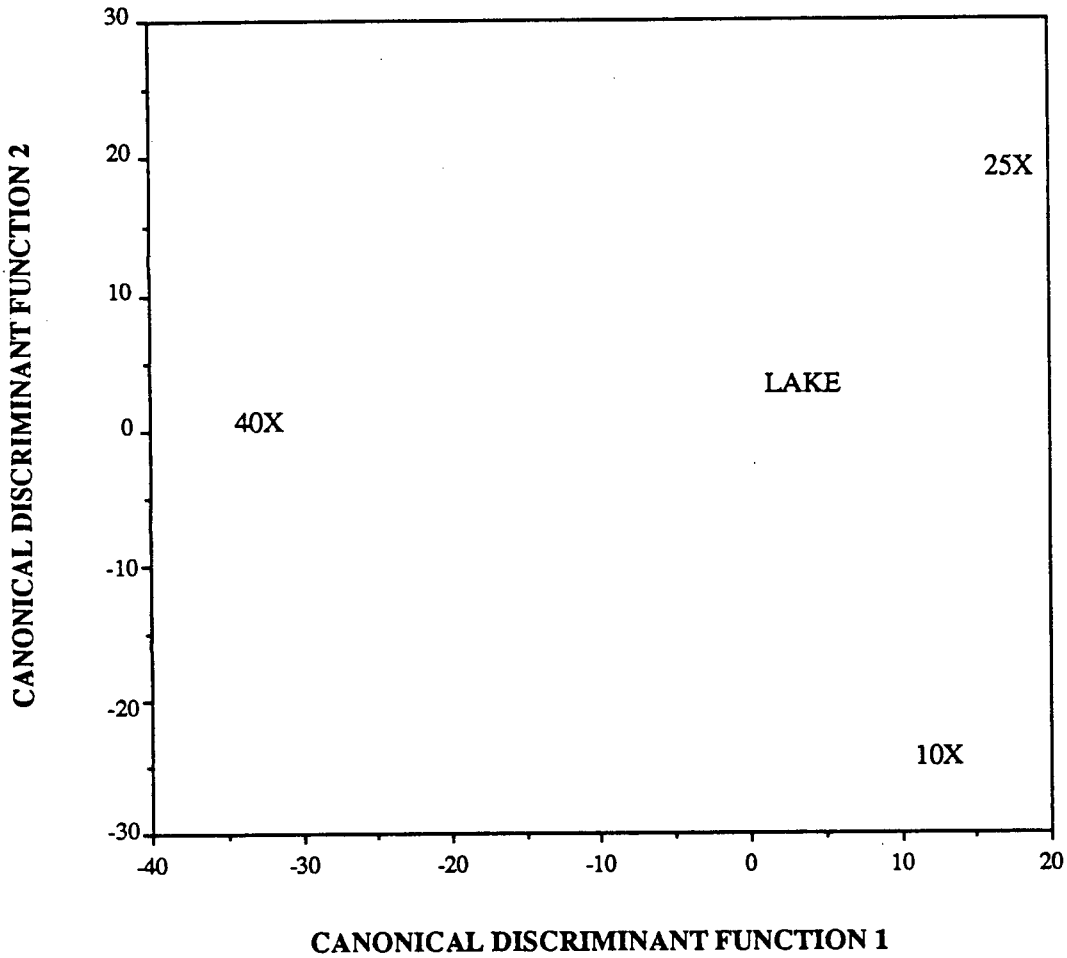


FIGURE 20: Canonical discriminant function scores for phytoplankton community composition (lake and "nutrient" enclosures).

TABLE XVIII: Summary table of discriminant analysis of phytoplankton composition in the lake and enclosures, with minimized Wilks' lambda.

SUMMARY TABLE

Step	Entered	Wilks' Lambda	P <
1	*Ankistrodesmus*	0.0061	0.001
2	*Selenastrum*	0.0001	0.001
3	Cryptomonads	0.0001	0.001
4	*Glenodinium*	0.0000	0.001
5	*Crucigenia*	0.0000	0.001

CANONICAL DISCRIMINANT FUNCTIONS

Fxn.	Eigen Value	Perc. Var.	Cum. Var.	Canonical Correl.	Wilks' Lambda	X^2	df	P <
0					0.0000	102.6	15	0.001
1	508.83	62.19	62.19	0.999	0.0001	62.1	8	0.001
2	255.80	31.26	93.45	0.998	0.0183	26.0	3	0.001
3	53.61	6.55	100.00	0.991				

CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS

	FUNC 1	FUNC 2	FUNC 3
Crucigenia	-2.244	-1.340	-0.991
Selenastrum	0.731	0.996	1.082
Ankistrodesmus	2.782	1.721	0.509
Glenodinium	3.630	-4.295	-0.216
Cryptomonads	-4.136	4.196	0.057

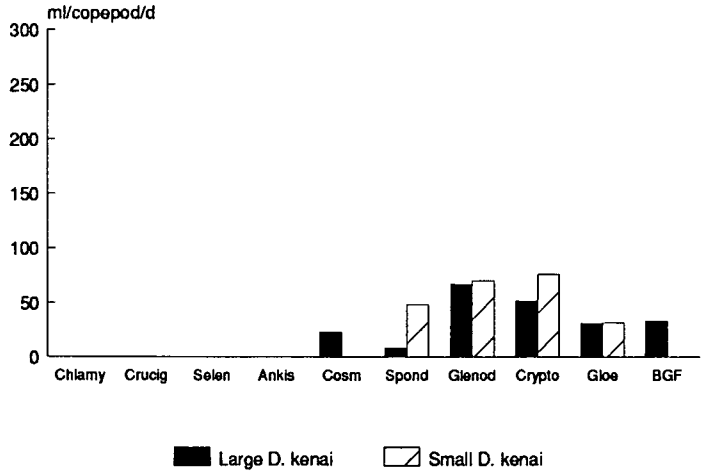
composition of large and small *D. kenai* indicated that the two copepods had similar diets, with similarity values ranging between 88% (feeding on lake and 10X phytoplankton communities) to 98% (feeding on 40X community). Comparisons of phytoplankton composition with diet composition yielded percent similarity values of 10% (25X community), 69% (lake), 96% (10X), and 98% (40X) for large *D. kenai* and 10% (25X community), 78% (lake), 83% (10X), and 97% (40X) for small *D. kenai*. The low percent similarity between diet composition and community composition for large and small *D. kenai* feeding on the 25X community is attributable to the high proportion of *Chlamydomonas* in the diet (63% and 50% for large and small *D. kenai*, respectively) compared to what was available in the phytoplankton community (4%).

Filtering rates of large and small *D. kenai* feeding on the different phytoplankton communities are shown in Figure 21. Hierarchical analysis (Figure 22) revealed that large and small *D. kenai* feeding on the 10X community had similar diets and these were joined with large *D. kenai* feeding on the 40X community. The FR values of large and small *D. kenai* feeding on the lake community were grouped at level 3. FR of small *D. kenai* feeding on the 40X community did not link with the other groups until level 15, indicating relatively high dissimilarity. The highest dissimilarity grouping linked a single FR function, one replicate of large *D. kenai* feeding on 25X phytoplankton, as most dissimilar from all other groups. Discriminant analysis separated FR data for large and small *D. kenai* and the four phytoplankton communities on the first two discriminant functions as shown in Figure 23. Nearly 87% of the total variance in diet composition was explained by the first two functions (Table XIX). The forward entry algorithm to minimize Wilks' lambda identified all phytoplankton taxa except *Gloetheca* as important in separating the groups.

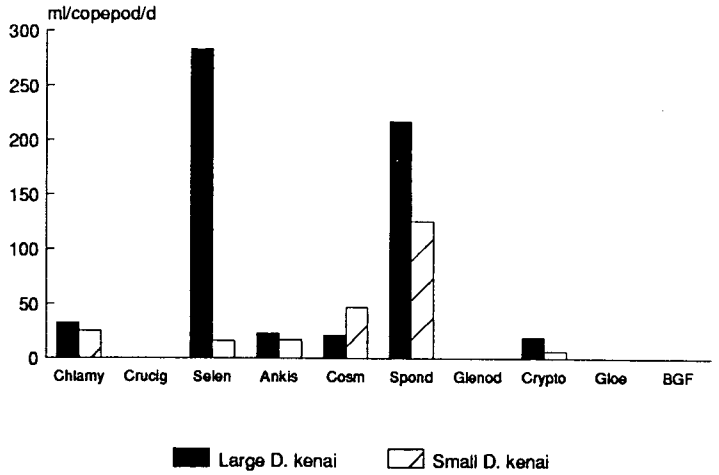
Figure 24 shows the E^* values observed for large and small *D. kenai* feeding on the different phytoplankton communities. For both large and small *D. kenai*, E^* values varied widely both among taxa as well as among enclosures. Only two taxa, *Crucigenia* and *Ankistrodesmus*, were

FIGURE 21: Filtering rate values for large and small *D. kenai* feeding on the lake and enclosure phytoplankton communities. (Chlamy: *Chlamydomonas*; Crucig: *Crucigenia*; Selen: *Selenastrum*; Ankist: *Ankistrodesmus*; Cosmar: *Cosmarium*; Spond: *Spondylosium*; Gleno: *Glenodinium*; Crypto: cryptomonads; Gloe: *Gloeotheca*; BGF: filamentous cyanobacteria)

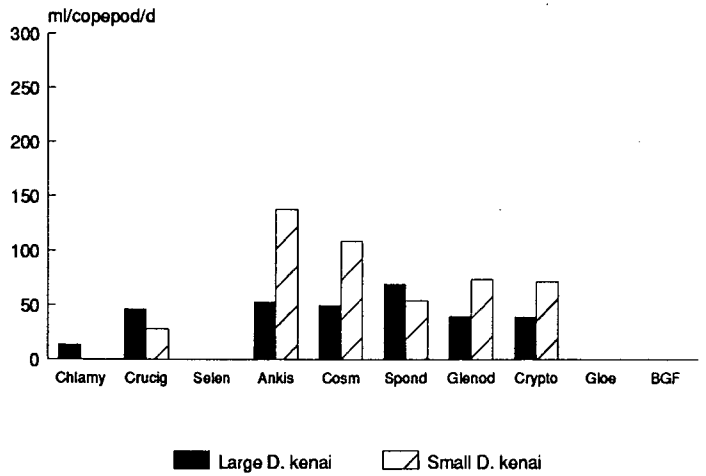
10X Enclosure



25X Enclosure



40X Enclosure



Lake

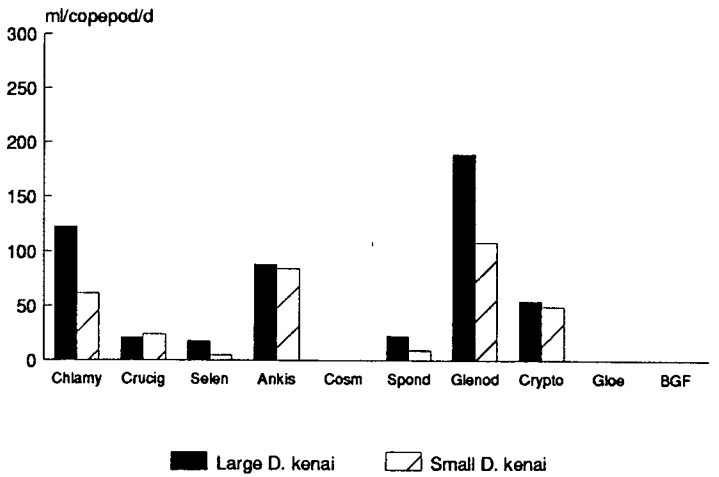
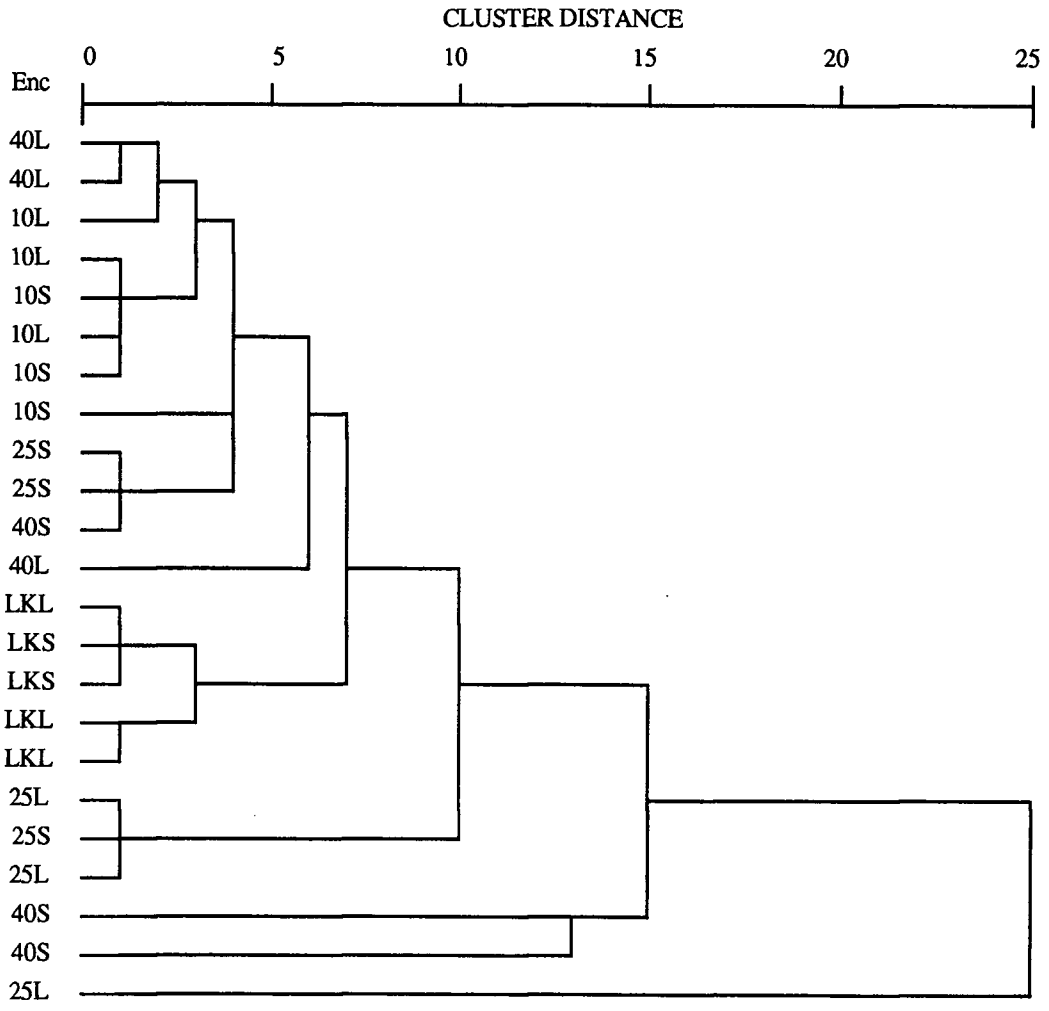


FIGURE 22: Dendrogram of hierarchical classification of filtering rates (FR) for large and small *Diaptomus kenai* feeding on the lake and enclosure phytoplankton communities. (10L: Large *D. kenai* feeding on 10X phytoplankton; 10S: Small *D. kenai* feeding on 10X phytoplankton; 25L: Large *D. kenai* feeding on 25X phytoplankton; 25S: Small *D. kenai* feeding on 25X phytoplankton; 40L: Large *D. kenai* feeding on 40X phytoplankton; 40S: Small *D. kenai* feeding on 40X phytoplankton; LKL: Large *D. kenai* feeding on Lake phytoplankton; LKS: Small *D. kenai* feeding on Lake phytoplankton)


CLUSTER DISTANCE
0
5
10
15
20
25
Enc
40L
40L
10L
10L
10S
10L
10S
10S
25S
25S
40S
40L
LKL
LKS
LKS
LKL
LKL
25L
25S
25L
40S
40S
25L

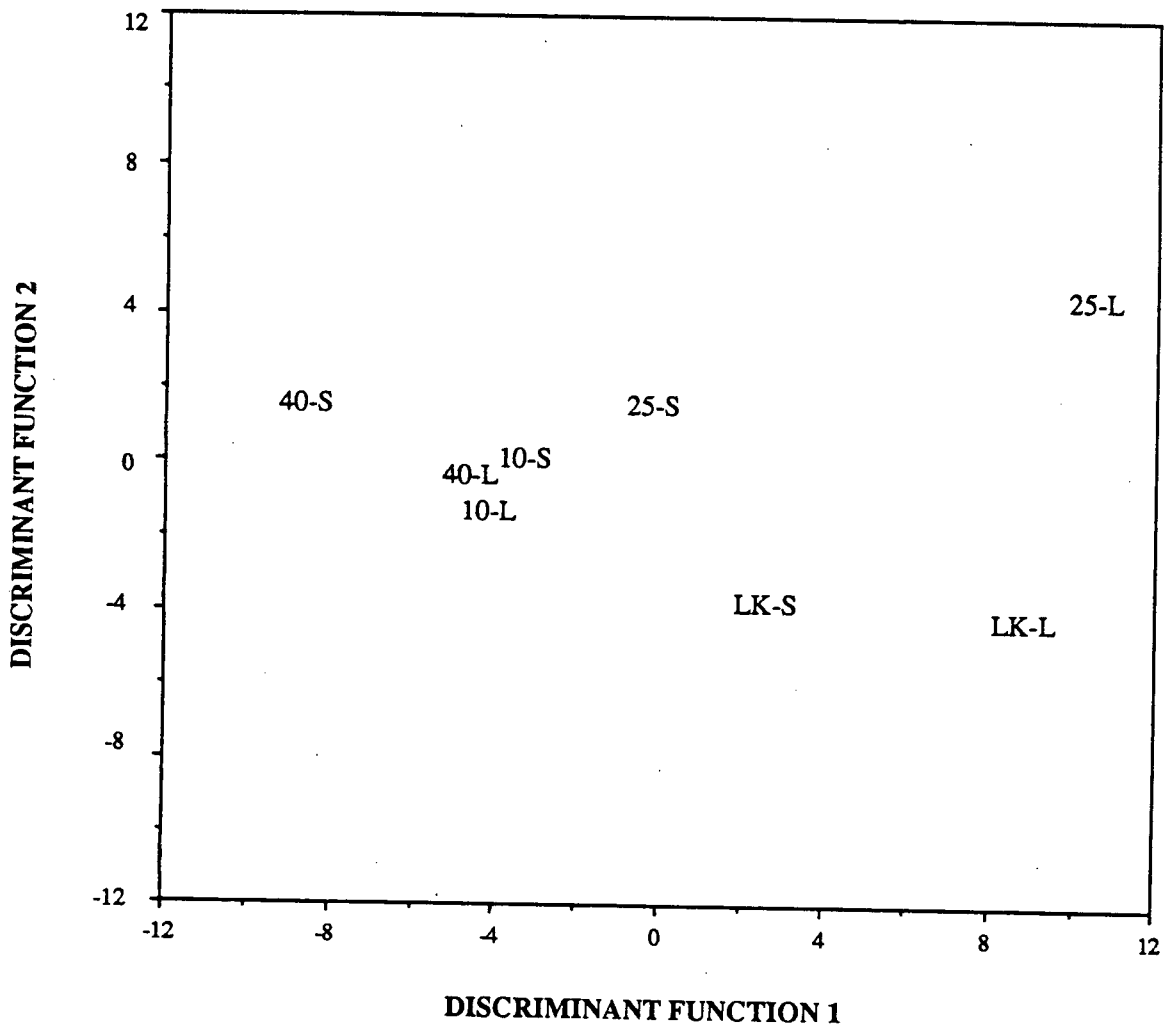


FIGURE 23: Canonical discriminant function scores of filtering rates (FR) for large and small *Diaptomus kenai* feeding on lake and enclosure phytoplankton communities. (See Figure 22 for definition of symbols)

TABLE XIX: Summary table of discriminant analysis of filtering rate (FR) values for large and small *Diaptomus kenai* feeding on phytoplankton from the lake and enclosures, with minimized Wilks' lambda.

SUMMARY TABLE

Step	Entered	Wilks' Lambda	P <
1	*Chlamydomonas*	0.0615	0.001
2	*Selenastrum*	0.0052	0.001
3	Filamentous Cyanobacteria	0.0020	0.001
4	*Glenodinium*	0.0008	0.001
5	*Cosmarium*	0.0003	0.001
6	*Crucigenia*	0.0001	0.001
7	*Ankistrodesmus*	0.0001	0.001
8	*Spondilosium*	0.0001	0.001

CANONICAL DISCRIMINANT FUNCTIONS

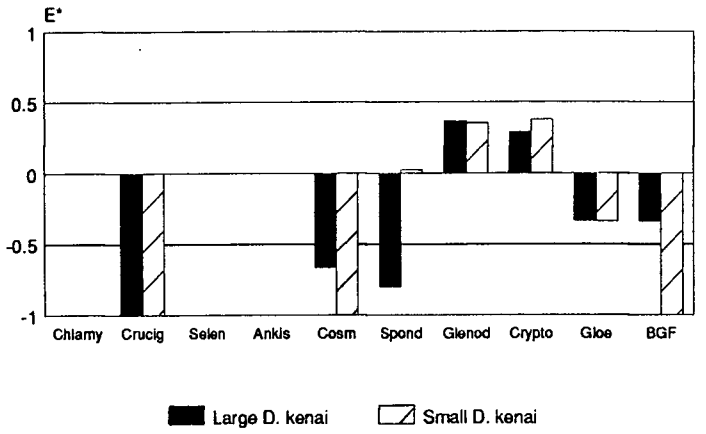
Fxn.	Eigen Value	Perc. Var.	Cum. Var.	Canonical Correl.	Wilks' Lambda	X^2	df	P <
0					0.0000	154.53	56	0.001
1	65.32	74.46	74.46	0.992	0.0011	95.80	42	0.001
2	10.92	12.45	86.91	0.957	0.0127	61.11	30	0.001
3	8.26	9.41	96.32	0.944	0.1177	30.00	20	0.07
4	1.67	1.91	98.23	0.792	0.3152	16.17	12	0.18
5	0.86	0.98	99.21	0.681	0.5870	7.46	6	0.28
6	0.67	0.76	99.98	0.633	0.9797	0.29	2	0.87
7	0.02	0.02	100.00	0.143				

CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS

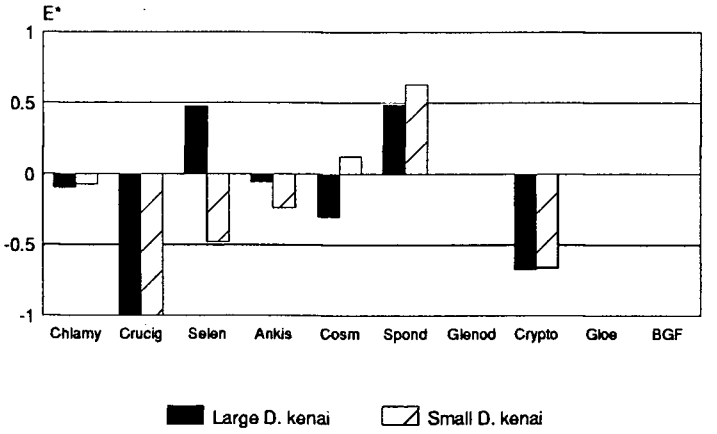
	FUNC 1	FUNC 2	FUNC 3
Chlamydomonas	1.392	0.007	0.602
Crucigenia	-0.978	1.101	1.517
Selenastrum	1.375	0.062	-0.440
Ankistrodesmus	0.422	-1.105	-0.379
Cosmarium	-1.427	0.813	1.528
Spondilosium	0.318	1.008	0.439
Glenodinium	-0.707	-0.478	-0.120
Filamentous Cyanobacteria	-0.141	-0.400	-0.510

FIGURE 24: Electivity indices for large and small *D. kenai* feeding on the lake and enclosure phytoplankton communities. (Chlamy: *Chlamydomonas*; Crucig: *Crucigenia*; Selen: *Selenastrum*; Ankist: *Ankistrodesmus*; Cosmar: *Cosmarium*; Spond: *Spondylosium*; Gleno: *Glenodinium*; Crypto: cryptomonads; Gloe: *Gloeotheca*; BGF: filamentous cyanobacteria)

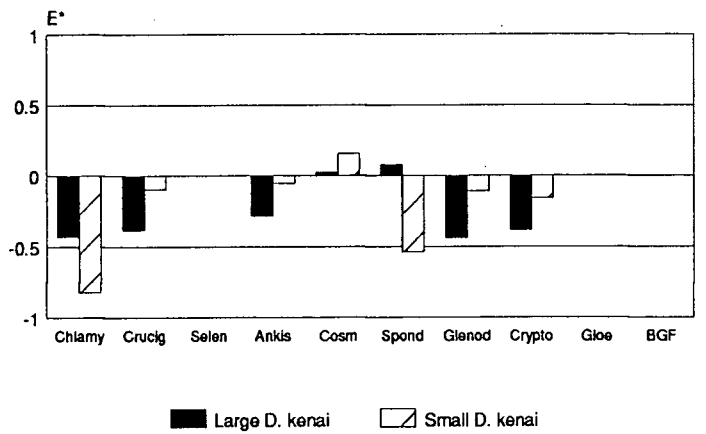
10X Enclosure



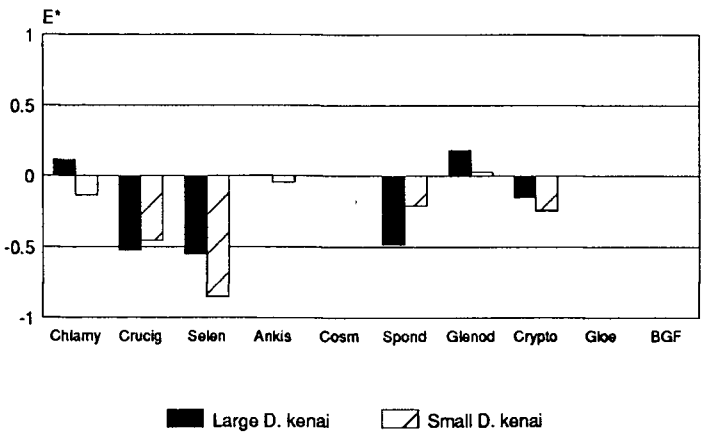
25X Enclosure



40X Enclosure



Lake



consistently avoided ($E^* < 0$) across all food treatments. E^* values for the remaining species ranged from avoided ($E^* < 0$) to preferred ($E^* < 0$) among the food treatments.

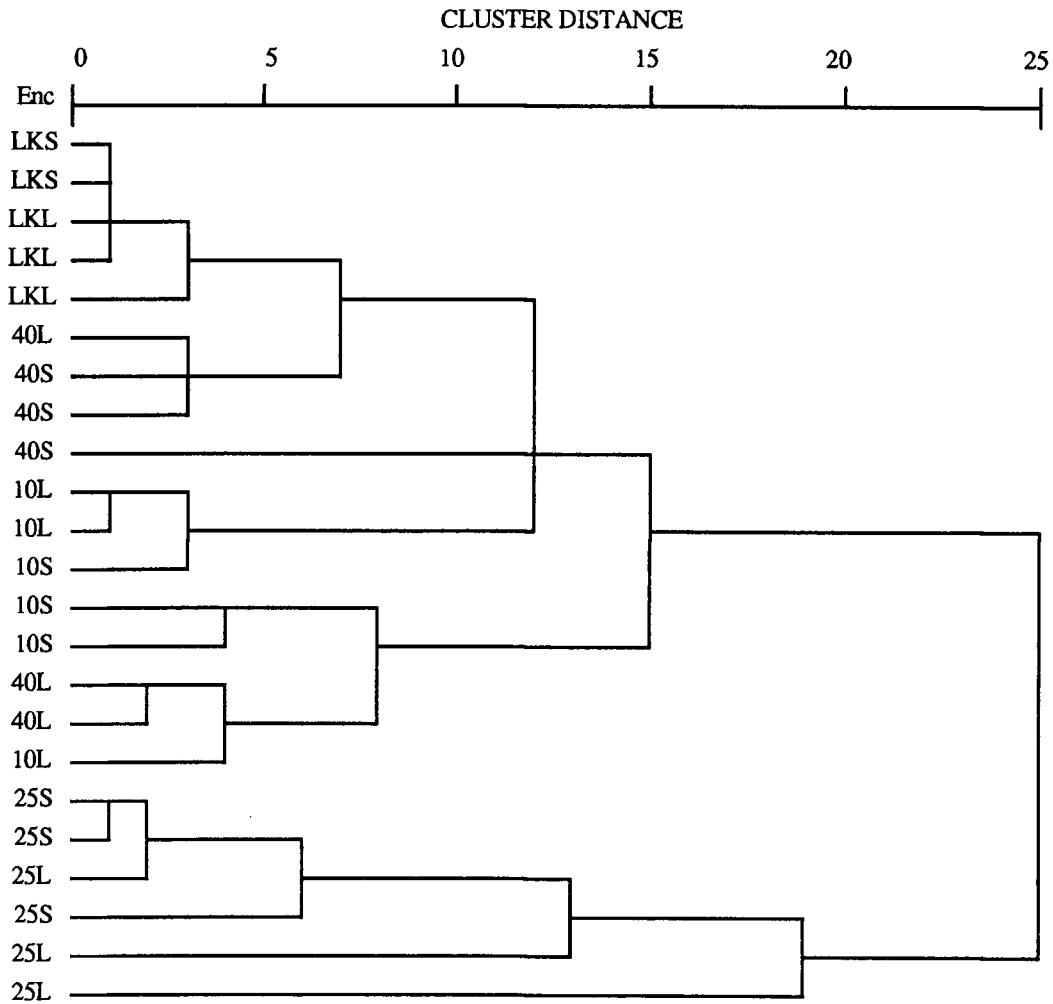
Hierarchical analysis of E^* values for large and small *D. kenai* feeding on the lake and enclosure phytoplankton communities (Figure 25) indicated that electivities for phytoplankton in the 25X enclosure were most different from all other groups, with a dissimilarity index of 25. Data from the lake community were grouped at level 3, with no discernible difference between large and small *D. kenai*. The remaining treatments did not pair off according to replicate, enclosure, or copepod, indicating no pattern of dissimilarity attributable to phytoplankton community or grazer.

While hierarchical analysis could not group the E^* values according to phytoplankton community or copepod, the eight groups were separated distinctly along the first and second discriminant functions (Figure 26). In this ordination of the complete data set, the first and second functions describe between them 94% of the variation (Table XX). The forward entry algorithm to minimize Wilks' lambda identified all phytoplankton taxa except *Gloetheca* as important in separating the groups.

Feeding Behavior and Food Density

Electivity indices and filtering rates of large and small *D. kenai* feeding on the different densities of the 40X phytoplankton community are presented in Figures 27 and 28. Large *D. kenai* filtering rate was higher at inermediate food densities, while small *D. kenai* filtering rate was higher at higher food densities. There was no systematic change in E^* indices for either large or small *D. kenai* as food density increased.

FIGURE 25: Dendrogram of hierarchical classification of electivity indices (E^*) of large and small *Diaptomus kenai* feeding on the lake and enclosure phytoplankton communities. (See Figure 22 for definition of symbols)


CLUSTER DISTANCE
0
5
10
15
20
25
Enc
LKS
LKS
LKL
LKL
LKL
40L
40S
40S
40S
10L
10L
10S
10S
10S
40L
40L
10L
25S
25S
25L
25S
25L
25L

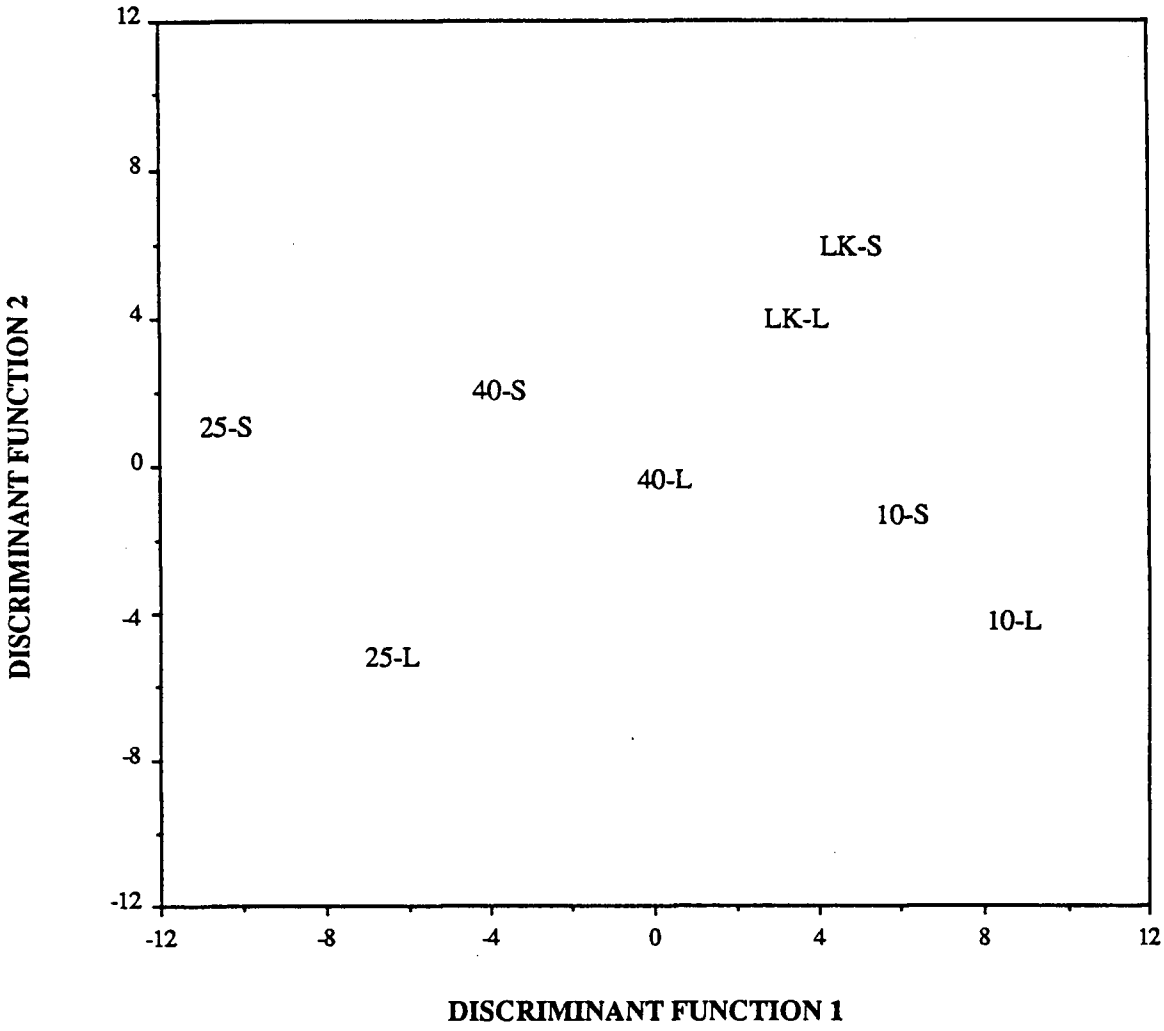


FIGURE 26: Canonical discriminant function scores of electivity indices (E^*) for large and small *Diaptomus kenai* feeding on lake and enclosure phytoplankton communities. (See Figure 22 for definition of symbols)

TABLE XX: Summary table of discriminant analysis of electivity indices (E^*) for large and small *Diaptomus kenai* feeding on phytoplankton from the lake and enclosures, with minimized Wilks' lambda.

SUMMARY TABLE

Step	Entered	Wilks' Lambda	P <
1	*Cosmarium*	0.1873	0.001
2	*Selenastrum*	0.0365	0.001
3	Cryptomonads	0.0074	0.001
4	*Glenodinium*	0.0026	0.001
5	Filamentous Cyanobacteria	0.0007	0.001
6	*Chlamydomonas*	0.0003	0.001
7	*Crucigenia*	0.0001	0.001
8	*Gloeotheca*	0.0001	0.001

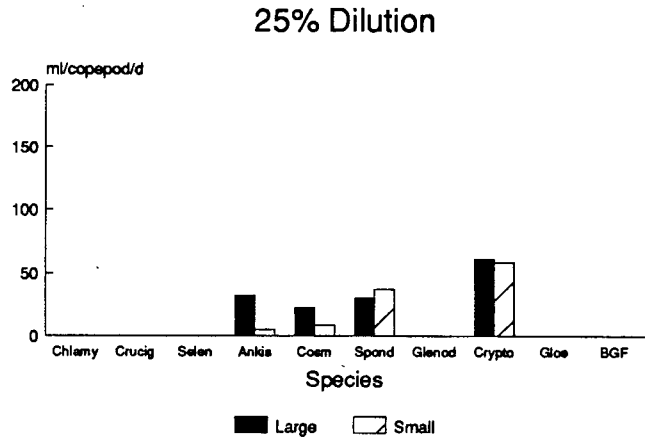
CANONICAL DISCRIMINANT FUNCTIONS

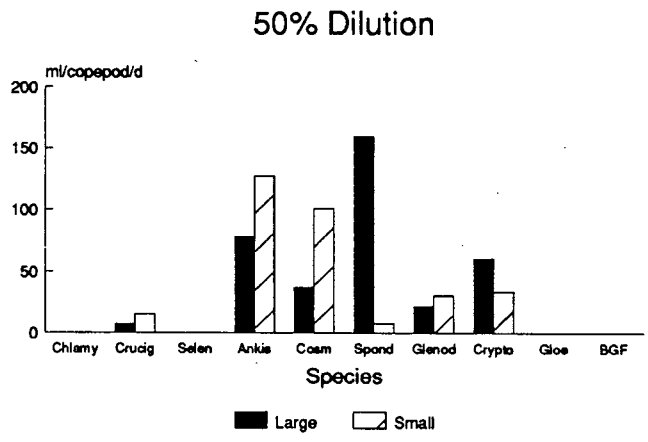
Fxn.	Eigen Value	Perc. Var.	Cum. Var.	Canonical Correl.	Wilks' Lambda	X^2	df	P <
0					0.000	136.95	56	0.001
1	60.51	72.16	72.16	0.992	0.004	79.28	42	0.001
2	18.67	22.26	94.42	0.974	0.068	37.57	30	0.16
3	2.45	2.93	97.35	0.843	0.236	20.22	20	0.44
4	1.69	2.01	99.36	0.792	0.633	6.39	12	0.90
5	0.45	0.54	99.90	0.557	0.919	1.19	6	0.98
6	0.07	0.08	99.98	0.256	0.983	0.24	2	0.89
7	0.02	0.02	100.00	0.130				

CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS

	FUNC 1	FUNC 2	FUNC 3
Chlamydomonas	1.432	1.050	-0.214
Crucigenia	0.741	1.377	-0.695
Selenastrum	-0.197	-0.919	-0.155
Cosmarium	-1.625	1.257	-0.388
Glenodinium	1.680	-0.009	0.249
Cryptomonads	2.368	0.012	0.333
Gloeotheca	-0.360	0.988	-0.741
Filamentous Cyanobacteria	1.027	-0.963	1.670

FIGURE 27: Filtering rates for large and small *D. kenai* feeding on different densities of the 40X enclosure phytoplankton community. (Chlamy: *Chlamydomonas*; Crucig: *Crucigenia*; Selen: *Selenastrum*; Ankist: *Ankistrodesmus*; Cosmar: *Cosmarium*; Spond: *Spondylosium*; Gleno: *Glenodinium*; Crypto: cryptomonads; Gloe: *Gloeotheca*; BGF: filamentous cyanobacteria)


25% Dilution
ml/copepod/d
200
150
100
50
0
Chlamy
Crucig
Selen
Ankis
Cosm
Spond
Glenod
Crypto
Gloe
BGF
Species
Large
Small


50% Dilution
ml/copepod/d
200
150
100
50
0
Chlamy
Crucig
Selen
Ankis
Cosm
Spond
Glenod
Crypto
Gloe
BGF
Species
Large
Small

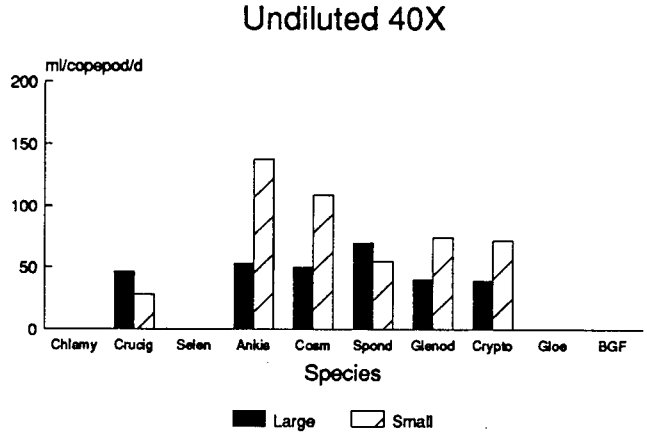
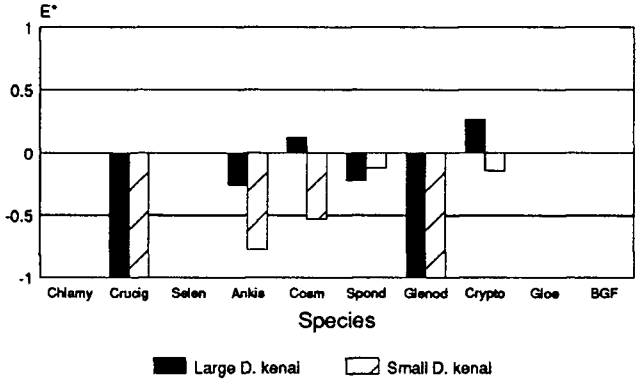
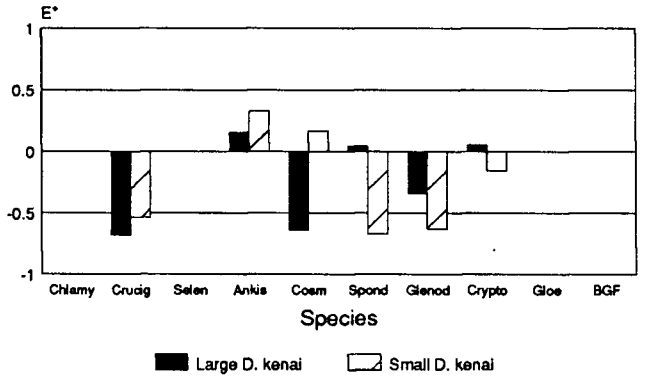

Undiluted 40X
ml/copepod/d
200
150
100
50
0
Chlamy
Crucig
Selen
Ankis
Cosm
Spond
Glenod
Crypto
Gloe
BGF
Species
Large
Small

FIGURE 28: Electivity indices for large and small *D. kenai* feeding on different densities of the 40X enclosure phytoplankton community. (Chlamy: *Chlamydomonas*; Crucig: *Crucigenia*; Selen: *Selenastrum*; Ankist: *Ankistrodesmus*; Cosmar: *Cosmarium*; Spond: *Spondylosium*; Gleno: *Glenodinium*; Crypto: cryptomonads; Gloe: *Gloeotheca*; BGF: filamentous cyanobacteria)

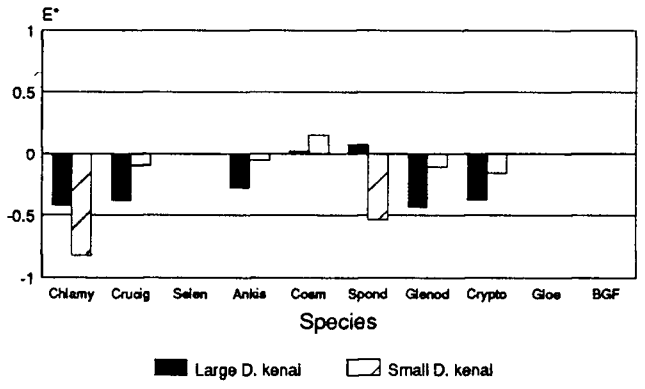
25% Dilution



50% Dilution



Undiluted 40X



Hierarchical grouping of FR data (Figure 29) did not produce any discernible pattern attributable to copepod size or food density except for the grouping of the replicates of one combination (small *D. kenai* feeding at the lowest food density). Discriminant analysis, however, did separate the groups along the first two discriminant functions (Figure 30), which together accounted for 87% of the total variation in FR (Table XXI).

Hierarchical grouping of E^* data linked groups according to food density, but not necessarily according to copepod size (Figure 31). Ordination of the complete data set on the first two discriminant functions grouped the data according to copepod size and food density (Figure 32). The first two functions accounted for 94% of the total variation in E^* among groups (Table XXII), with four taxa (*Glenodinium*, cryptomonads, *Spondylosium*, and *Ankistrodesmus*) being most important in explaining variation.

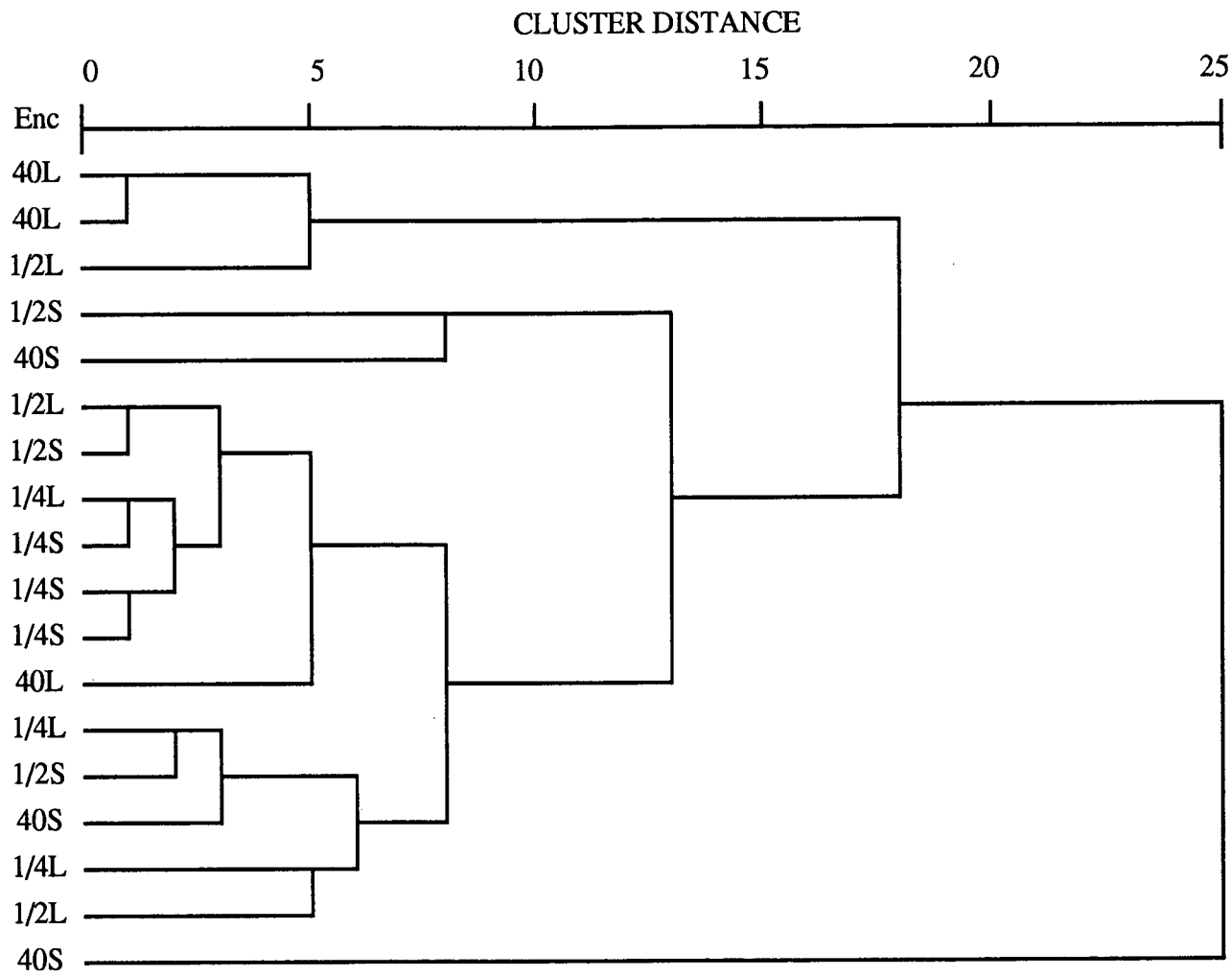
Sources of Variation in Feeding Behavior

To identify characteristics of individual phytoplankton taxa that might help to explain observed variations, FR and E^* were analysed for large and small *D. kenai* feeding on the lake phytoplankton community alone.

One-way ANOVA of FR and E^* values for large and small *D. kenai* feeding on the lake phytoplankton community indicated a significant taxon effect on FR (Table XXIII) but not E^* (Table XXIV). Regression analysis of algal attributes which might contribute to a taxa effect could not identify a significant linear relationship with any of the attributes examined (GALD, volume, individual taxon concentration, total concentration, percent of community).

Multiple regression analysis of feeding on each phytoplankton taxon, compared among the four phytoplankton communities (Table XXV), indicated FR and E^* were linearly associated with different independent variables according to the size of copepod and the taxon analyzed; that is, there was no single variable by which feeding behavior varied consistently

FIGURE 29: Dendrogram of hierarchical classification of filtering rates (FR) large and small *Diaptomus kenai* feeding at different food densities. (1/4L: Large *D. kenai* feeding on 1/4 concentration 40X phytoplankton; 1/4S: Small *D. kenai* feeding on 1/4 concentration 40X phytoplankton; 1/2L: Large *D. kenai* feeding on 1/2 concentration 40X phytoplankton; 1/2S: Small *D. kenai* feeding on 1/2 concentration 40X phytoplankton; 40L: Large *D. kenai* feeding on undiluted 40X phytoplankton; 40S: Small *D. kenai* feeding on undiluted 40X phytoplankton)


CLUSTER DISTANCE
0
5
10
15
20
25
Enc
40L
40L
1/2L
1/2S
40S
1/2L
1/2S
1/4L
1/4S
1/4S
1/4S
40L
1/4L
1/2S
40S
1/4L
1/2L
40S

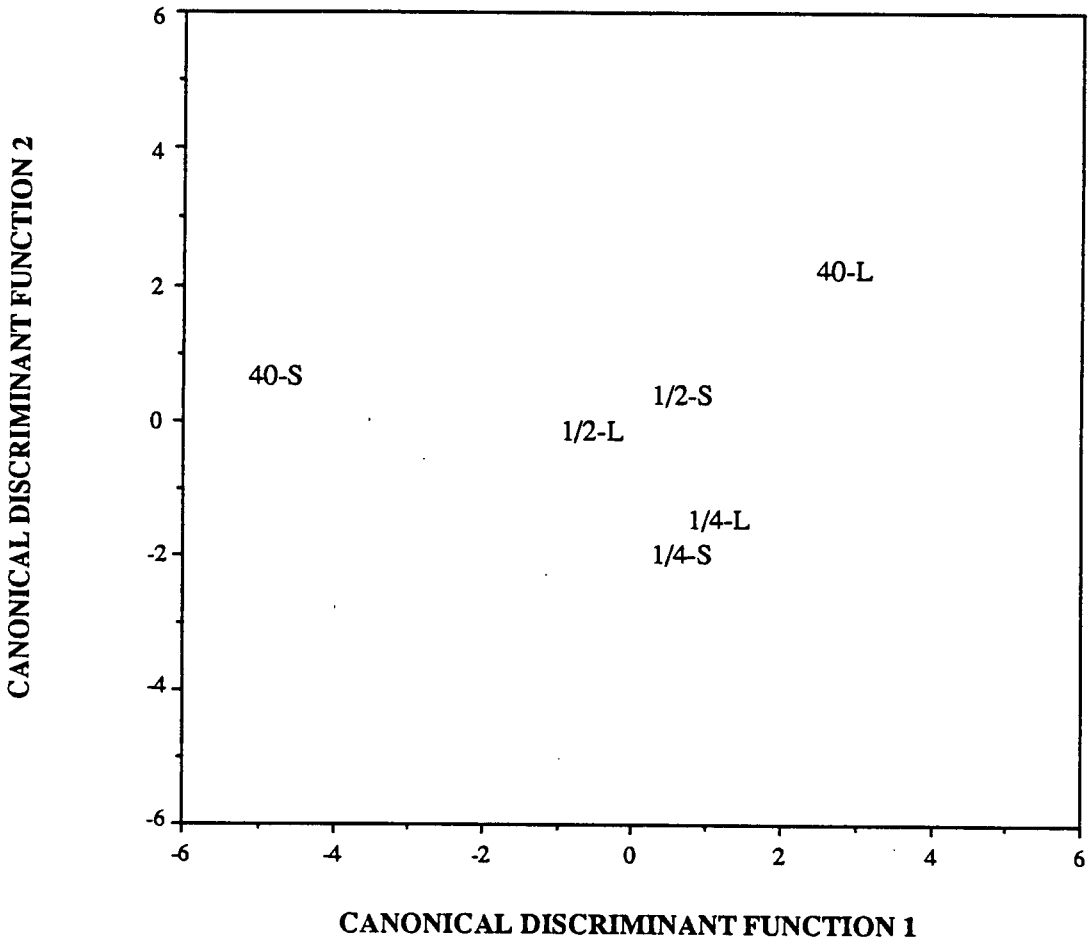


FIGURE 30: Canonical discriminant function scores of filtering rates (FR) for large and small *Diaptomus kenai* feeding at different phytoplankton densities. (See Figure 29 for definition of symbols)

TABLE XXI: Summary table of discriminant analysis of filtering rate (FR) values for large and small *Diaptomus kenai* feeding on different densities of phytoplankton, with minimized Wilks' lambda.

SUMMARY TABLE

Step	Entered	Wilks' Lambda	P <
1	*Glenodinium*	0.3171	0.01
2	*Spondilosium*	0.1639	0.01
3	*Crucigenia*	0.0524	0.002
4	Cryptomonads	0.0255	0.002
5	*Cosmarium*	0.0136	0.004
6	*Ankistrodesmus*	0.0077	0.01

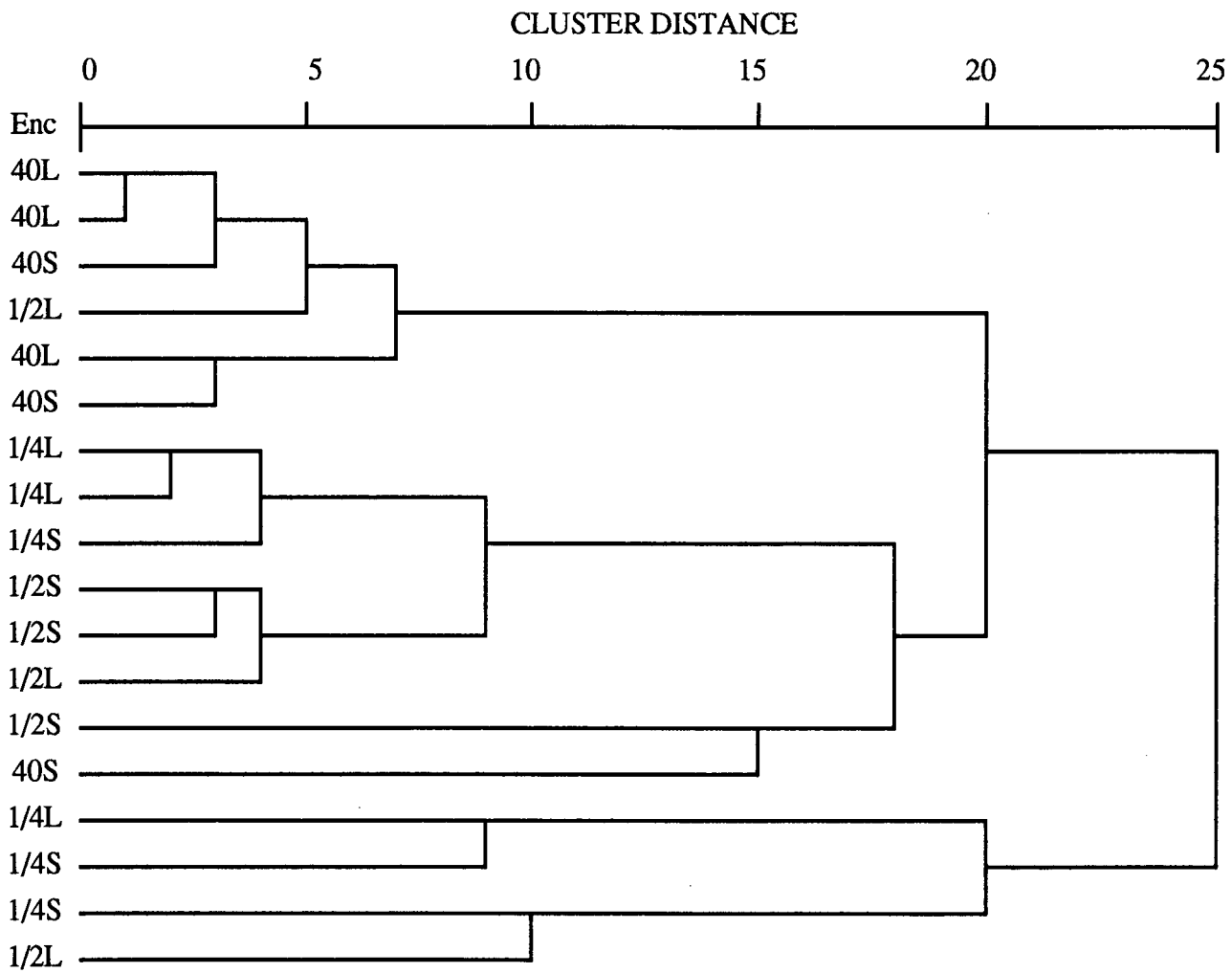
CANONICAL DISCRIMINANT FUNCTIONS

Fxn.	Eigen Value	Perc. Var.	Cum. Var.	Canonical Correl.	Wilks' Lambda	X^2	df	P <
0					0.008	53.469	30	0.005
1	8.32	63.26	63.26	0.945	0.072	28.914	20	0.09
2	3.15	23.96	87.22	0.871	0.300	13.256	12	0.35
3	1.20	9.12	96.34	0.738	0.659	4.589	6	0.60
4	0.39	2.97	99.31	0.530	0.916	0.962	2	0.62
5	0.09	0.69	100.00	0.289				

CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS

	FUNC 1	FUNC 2	FUNC 3
Glenodinium	2.698	1.167	0.210
Crucigenia	0.071	0.466	0.723
Cryptomonads	-0.427	-0.065	0.661
Cosmarium	1.764	0.728	-0.026
Ankostrodesmus	-3.148	-0.014	-0.663
Spondilosium	0.043	-1.196	-0.301

FIGURE 31: Dendrogram of hierarchical classification of electivity indices (E^*) large and small *Diaptomus kenai* feeding at different food densities. (See Figure 29 for explanation of symbols)


CLUSTER DISTANCE
0
5
10
15
20
25
Enc
40L
40L
40S
1/2L
40L
40S
1/4L
1/4L
1/4S
1/2S
1/2S
1/2L
1/2S
40S
1/4L
1/4S
1/4S
1/2L

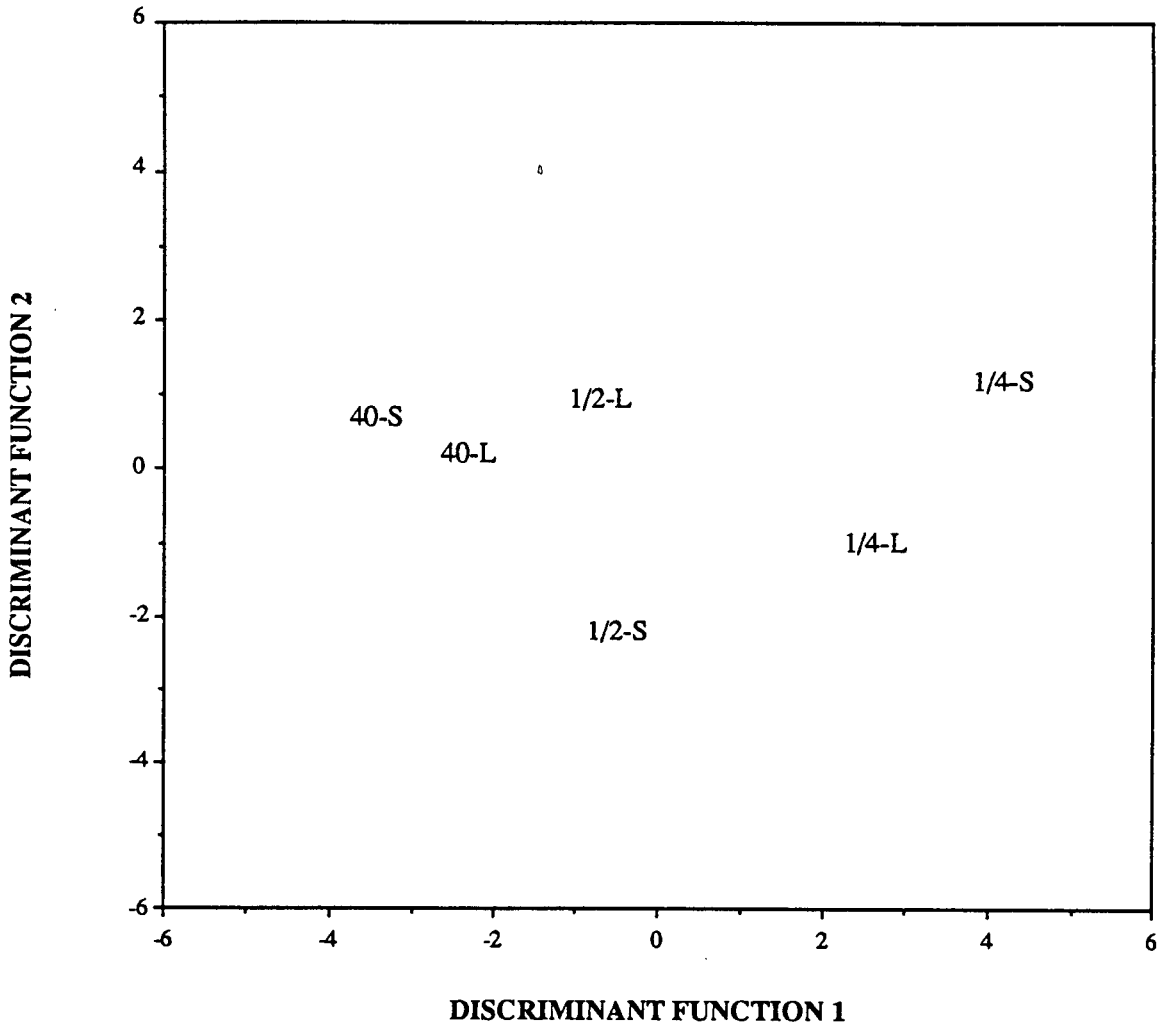


FIGURE 32: Canonical discriminant function scores of electivity indices (E*) for large and small *Diaptomus kenai* feeding at different phytoplankton densities. (See Figure 29 for definition of symbols)

TABLE XXII: Summary table of discriminant analysis of electivity indices (E^*) for large and small *Diaptomus kenai* feeding on different densities of phytoplankton, with minimized Wilks' lambda.

SUMMARY TABLE

Step	Entered	Wilks' Lambda	P <
1	*Glenodinium*	0.1768	0.001
2	Cryptomonads	0.0562	0.001
3	*Sponilosium*	0.0268	0.001
4	*Ankistrodesmus*	0.0143	0.001

CANONICAL DISCRIMINANT FUNCTIONS

Fxn.	Eigen Value	Perc. Var.	Cum. Var.	Canonical Correl.	Wilks' Lambda	X^2	df	P <
0					0.014	50.948	20	0.001
1	10.20	77.32	77.32	0.954	0.161	21.956	12	0.04
2	2.15	16.28	93.60	0.826	0.505	8.194	6	0.22
3	0.63	4.78	98.38	0.622	0.824	2.322	2	0.31
4	0.21	1.62	100.00	0.419				

CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS

	FUNC 1	FUNC 2	FUNC 3
Ankostrodesmus	-0.340	0.440	1.063
Spondilosium	0.733	0.525	0.317
Glenodinium	-0.818	0.840	-0.077
Cryptomonads	0.704	1.075	-0.239

TABLE XXIII: One-way analysis of variance of a)large *Diaptomus kenai* and b) small *D. kenai* filtering rate with phytoplankton taxon as the independent variable.

a) large *Diaptomus kenai*

SOURCE	SS	DF	F	P <	R^2
Model	106838.9	6	8.404	0.001	0.783
Residual	29662.0	14			

a) small *Diaptomus kenai*

SOURCE	SS	DF	F	P <	R^2
Model	33946.5	6	7.826	0.001	0.770
Residual	10120.8	14			

TABLE XXIV: One-way analysis of variance of a)large *Diaptomus kenai* and b) small *D. kenai* electivity indices with phytoplankton taxon as the independent variable.

a) large *Diaptomus kenai*

SOURCE	SS	DF	F	P <	R^2
Model	1.792	6	1.951	0.15	0.455
Residual	2.143	14			

a) small *Diaptomus kenai*

SOURCE	SS	DF	F	P <	R^2
Model	1.593	6	1.217	0.35	0.343
Residual	3.054	14			

TABLE XXV: Stepwise multiple regression statistics describing the effect of total cell concentration ([T]), taxon concentration ([tax]), enclosure (enc), and species diversity (#spp) on large and small *Diaptomus kenai* a) electivity indices and b) filtering rates based on data from the feeding experiments using phytoplankton communities from the lake and enclosures.

		a) Electivity Indices			**b) Filtering Rates**		
Taxon	Copepod	Regression Equation (Standardized Coefficients)	R^2	P <	Regression Equation (Standardized Coefficients)	R^2	P <
Chlamy	large	E* = -0.743(enc)	0.553	0.05	FR = -0.963(enc)	0.928	0.001
	small	E* = 0.813([tax])	0.662	0.01	FR = -0.930(enc)	0.865	0.001
Cruc	large	E* = -0.613(#spp)	0.376	0.05	FR = n.s.		
	small	E* = -0.619(#spp) - 0.520([T])	0.832	0.001	FR = -0.681([T])	0.464	0.01
Sel	large	E* = 1.95(#spp) + 1.04([tax])	0.994	0.001	FR = n.s.		
	small	E* = n.s.			FR = n.s.		
Ank	large	E* = n.s.			FR = -0.740([T])	0.548	0.05
	small	E* = n.s.			FR = n.s.		
Cosm	large	E* = 0.711([T])	0.506	0.05	FR = -0.716([T])	0.513	0.05
	small	E* = -1.02([T]) - 0.364([tax])	0.933	0.001	FR = -0.791([T])	0.625	0.05
Spond	large	E* = n.s.			FR = n.s.		
	small	E* = 0.700(#spp)	0.490	0.05	FR = n.s.		
Gleno	large	E* = 1.40(#spp) + 0.84([tax])	0.921	0.001	FR = -0.670(enc)	0.449	0.05
	small	E* = n.s.			FR = n.s.		
Crypto	large	E* = n.s.			FR = n.s.		
	small	E* = 0.768([T]) - 0.777(#spp) - 0.375(enc)	0.889	0.001	FR = -1.825(#spp) - 1.679([tax]) + 0.671(enc)	0.952	0.001

among the taxa. Neither was there a consistent pattern in factors affecting E^* for large and small *D. kenai* feeding on each taxon; factor(s) associated with E^* values of large *D. kenai* were not necessarily the same as those for small *D. kenai* feeding on the same taxon. E^* had a significant linear association with number of alternative food choices (i.e., variable "number of taxa") for five of the taxa examined for either one or both copepods. A positive β value indicates increased electivity for that taxon as the number of alternative food choices increases (e.g., small *D. kenai* feeding on *Spondylosium*), while a negative value indicates decreased electivity with an increase in alternative food choices (e.g., both large and small *D. kenai* feeding on *Crucigenia*). There was also a significant linear association between E^* and either food density and/or individual taxon density for six of the taxa analyzed. Enclosure effects were significant in two cases only: large *D. kenai* feeding on *Chlamydomonas* and small *D. kenai* feeding on cryptomonads. E^* on *Ankistrodesmus* was not associated with any of the variables tested for either copepod.

Two independent variables frequently emerged as being associated with FR among the various taxa (Table XXV). Enclosure was more frequently associated with FR than was E^*, and was for two taxa (*Chlamydomonas* and *Glenodinium*) the only significant independent variable. Total cell concentration was the only significant independent variable for three taxa (*Crucigenia*, *Ankistrodesmus*, and *Cosmarium*).

Analysis of feeding on each phytoplankton taxon compared among the three densities of the 40X phytoplankton community identified, in those few taxa where a significant linear association was indicated, dilution factor as one of the significant variables associated with FR and E^* in nearly every case (Table XXVI). However, for two taxa (*Cosmarium* and *Spondylosium*) neither FR nor E^* for either copepod were significantly associated with any of the independent variables examined. In addition, for only two of the six taxa studied was FR significantly associated with any of the independent variables (*Crucigenia* and *Glenodinium*).

TABLE XXVI: Stepwise multiple regression statistics describing the effect of total cell concentration ([T]), taxon concentration ([tax]), and dilution factor (dil) on large and small *Diaptomus kenai* a) electivity indices and b) filtering rates based on data from the feeding experiments using different dilutions of the 40X enclosure phytoplankton community

		a) Electivity Indices			**b) Filtering Rates**		
Taxon	Copepod	Regression Equation (Standardized Coefficients)	R^2	P <	Regression Equation (Standardized Coefficients)	R^2	P <
Cruc	large	E* = -0.913([tax])	0.833	0.001	FR = -0.831([tax])	0.690	0.01
	small	E* = -0.824(dil)	0.679	0.01	FR = -0.863(dil)	0.745	0.01
Ank	large	E* = -0.739([tax])	0.546	0.05	FR = n.s.		
	small	E* = n.s.			FR = n.s.		
Cosm	large	E* = n.s.			FR = n.s.		
	small	E* = n.s.			FR = n.s.		
Spond	large	E* = n.s.			FR = n.s.		
	small	E* = n.s.			FR = n.s.		
Gleno	large	E* = n.s.			FR = 0.675([T])	0.455	0.05
	small	E* = 0.749(dil)	0.561	0.05	FR = -0.719(dil)	0.516	0.05
Crypto	large	E* = 0.843(dil)	0.711	0.01	FR = n.s.		
	small	E* = 2.43(dil) + 8.73([T]) - 7.12([tax])	0.946	0.01	FR = n.s.		

DISCUSSION

The results presented here demonstrate that the two size classes of *Diaptomus kenai* have variable food-utilization patterns which differ between the two size classes as well as among different food regimes. This variablility is a flexible response which is manifest within 6 h of being placed in a new food assemblage, although it can be subtle in expression. This variability in feeding does not appear to be a response to any of the criteria typically acknowledged as important in influencing feeding behavior and the actual nature of the response varies according to how the food regime is varied. For example, in the study in which food composition is varied, hierarchical analysis consistently groups FR data according to treatment, while E^* values are linked randomly with respect to treatment. On the other hand, when food density is varied, E^* values are consistently grouped according to treatment, while FR values are grouped randomly.

No feeding pattern was consistently associated with either overall food density or density of that taxon as predicted in the early models of Solomon (1949) and Holling (1959) and demonstrated in zooplankton feeding studies (e.g., Richman 1966; Haney and Trout 1985; Stemberger 1986). Neither is feeding predictably associated with attributes such as cell size or cell shape as has been demonstrated for other copepods (e.g., McQueen 1970; O'Connors *et al.* 1980; Richman *et al.* 1980; Price and Paffenhöfer 1985; Chow-Fraser 1986; Vanderploeg 1990).

Most striking are the variations in electivity values within food taxa; that is, variation in electivity for a food among different food regimes. Cyanobacteria, which are typically avoided by most grazers, are ingested by both large and small *D. kenai*, with the exception being the total avoidance of filamentous forms by small *D. kenai*. Considering the substantial volume of literature providing evidence of the potential toxic properties (Porter and Orcutt 1980; Lampert 1981; Nizan *et al.* 1986; Threlkeld 1986) or interference effects (Hartman

1985; Hawkins and Lampert 1989; Gliwicz and Lampert 1990) of cyanobacteria as well as those which demonstrate that copepods avoid ingesting cyanobacteria altogether (Hartman 1985; Fulton and Paerl 1988; Vanderploeg *et al* 1990), these results are quite surprising. Even foods typically classified as nutritionally "good" were not consistently favored. For example, *Cryptomonas*, frequently used in feeding studies as a "high quality" food (Cowgill *et al.* 1985; Williamson *et al.* 1985; Williamson and Butler 1986; Fulton and Paerl 1987), is positively selected in only one of the four food treatments, with negative E^* values in the other three. For most algal taxa selectivity varied among the different food regimes; a food avoided ($E^* < 0$) under one condition could be preferred ($E^* > 0$) under another. Consistent electivities were observed for only two taxa (*Crucigenia* and *Ankistrodesmus*), which were always avoided.

These results are reminiscent of Conover's (1978) criticisms of laboratory feeding studies, in which he argues against the existence of satiation in nature. He concludes that it is inappropriate to ascribe phenomena observed in the laboratory under artificial feeding conditions to feeding in nature. In his criticisms he specifically addresses the use of excessively high food concentrations to demonstrate the existence of satiation, but similar criticisms can be applied to studies which attempt to describe feeding under natural food conditions with feeding models generated under very specific laboratory conditions. Feeding in nature involves decision making in a dilute, but compositionally complex, food regime. As my results demonstrate, feeding behavior in a complex food environment does not necessarily reflect behaviors observed in laboratory studies. Most laboratory feeding studies involve comparison of feeding on a small number of food types (usually only one or two different foods) offered alone or in combination, usually at unnaturally high (relative to normal, or field) concentrations. The criteria by which a copepod makes a foraging decision in laboratory studies, which, of necessity, include only a limited number of relevant environmental variables, may have only marginal bearing on the criteria by which food is ingested in a more complex food regime. Accordingly, the feeding behaviors observed in

this study could not be explained by laboratory-generated models typically used to describe and predict feeding in nature.

Exceptions to the overly simplistic, and overly concentrated, design are those feeding studies that assess selectivity based on food size. In these studies, copepods are offered a mixture of natural plankton and changes in phytoplankton size structure are identified using an electronic particle counter (e.g., Vanderploeg 1981; Vanderploeg *et al.* 1984). However, it has been pointed out that the utility of electivity based on food size is questionable when foraging decisions may be subject to modification based on such factors as food quality and food concentration (Pafferhöfer 1988). These results support those cautions, for, when comparing feeding among phytoplankton communities generated under different nutrient regimes, feeding behavior is independent of cell size and may be due more to the interactions of community structure and cell chemistry.

Not only do these foraging decisions vary according to characteristics of the food regime, but they also vary between the two size classes of *D. kenai*. Regardless of whether the food regime is varied by cell density or taxonomic composition, the two copepods (large and small *D. kenai*) appear to ingest food by different criteria. Investigators have demonstrated differences in feeding behavior among different species (Poulet and Oulet 1982; Daro 1985; DeMott 1988), but I am unaware of any work which demonstrates variation in selectivity within a single population as presented here, other than reports of ontogenetic changes in feeding (e.g., Paffenhöfer and Knowles 1978; Paffenhöfer 1984; Chow-Fraser and Wong 1986). In addition, these results support the findings in the previous chapter, which suggests that the food value of different phytoplankton communities to the copepods (as measured by copepod lipid stores) may vary between the two sizes.

Chapter Five

GENERAL DISCUSSION

Copepods live in what has been described as a nutritionally dilute environment (Conover 1968), experiencing a range of temporally as well as spatially variable food conditions which differ in their level of predictability. For example, migrating zooplankton experience spatially variable food on a predictable daily cycle. Seasonal variation in phytoplankton density and composition can be highly predictable in most temperate lakes. Intermediate between large scale seasonal variability and short scale daily variation are the less predictable, often chance, events (described as "event scale" variation by Landry and Hassett 1985). For example, sudden blooms and crashes of phytoplankton species or communities can occur on time scales of days or weeks, while wind-generated currents can create spatial heterogeneity in the phytoplankton on time scales of hours or days.

Understanding the mechanisms by which organisms deal with these changes in their food environment has been cited as a key problem in elucidating the nutritional ecology of zooplankton and the role of food limitation in structuring zooplankton communities (Landry and Hassett 1985). There is a considerable body of evidence that food resources limit zooplankton (Neill 1978, 1981a,b, 1985; Kerfoot and DeMott 1980; Neill and Peacock 1980; Frost 1985; Huntley 1985) and it is quite likely that food availability periodically falls below maintenance levels (Lampert 1977; Lampert and Schober 1980; Threlkheld 1986). The pronounced temporal variation in egg production observed in field populations of copepods (Edmondson *et al.* 1962; Comita 1964; Armitage *et al.* 1973; Kimmerer 1984; Smith and Lane 1985; Peterson 1985) also suggests that food levels periodically fail to meet energetic or nutrient needs. The strategies by which zooplankton respond to variation in food supply may be quite different depending upon the scale of the variation (i.e., daily, event, or seasonal) (Landry and Hassett 1985) and the differences among species in their perception of temporal

and spatial variation in food availability (Donaghay 1988).

In this thesis I examined changes in behavior, morphology, and physiology of two species of calanoid copepods in response to variation in food composition over a range of time scales and demonstrated that response flexibility varied depending upon the type of response measured. But the most striking finding in this study was the occurrence of two co-existing size classes of *Diaptomus kenai*. Morphological variation within a species is not a unique phenomenon; in fact, it has been well documented in a host of organisms including plants, invertebrates, and vertebrates. The recent review of Stearns (1989) discusses phenotypic flexibility within the framework of "reaction norm", or the relationship between a variable environment and a flexible phenotype that varies continuously with the environment. These reaction norms may be indirect responses to environmental gradients, such as increase in body size associated with decreased temperatures, commonly reported for insects, crustaceans, fish, amphibians, and reptiles. They may impart a distinct adaptive advantage ("adaptive reaction norm"), resulting in enhanced growth, survival, or reproduction, such as the changes in cichlid jaw morphology to adapt to changes in diet (Meyer 1987) or changes in cladoceran body size or shape to reduce predation risk (Dodson 1989). However, the observed variation in *Diaptomus kenai* size in Shirley Lake is not a temporally or spatially variable phenomenon that changes with any recognizable environmental cue; the two size classes co-occur, both throughout the lake and throughout the year.

Because the two size classes co-occur, the difference in size of *D. kenai* in Shirley Lake cannot be readily ascribed to some seasonal phenomenon. Therefore, there must be some advantage attributable to each size class that permits coexistence and persistence of two distinct, reproductively isolated phenotypes. It is quite possible that subtle differences in feeding behaviors and food requirements are sufficient to separate the two morphologically distinct subpopulations into two ecologically distinct subpopulations. It has been suggested that differences in body size permit calanoid copepod species to coexist, the theory being

that smaller-bodied copepods eat smaller sized particles than larger sized copepods, and competition for food resources is thereby minimized (Maly 1973, 1976; Maly and Maly 1974). Accordingly, Hutchinson (1967) proposed that a size difference of 35% (and Hammer and Sawchyn [1968] a difference of 0.5 mm or more, regardless of actual body size) would be sufficient to prevent food niche overlap between two co-existing diaptomid copepods. While these rules may apply to size differences between, and help explain the coexistence of, *D. kenai* and *D. leptopus* (mean body length 2.3 mm and 1.2 mm, respectively), they certainly do not apply to the relatively smaller difference between the two sizes of *D. kenai* (mean body length 2.0 and 2.3 mm).

The idea that different phytoplankton communities might be of different food value is supported by the pronounced differences observed among the enclosures in the amount of stored lipids accumulated by the copepods during the first year of my study, suggesting differences in the ability of the copepods to meet energetic demands when feeding on those phytoplankton communities. Further support for this idea is lent by the pronounced differences in clutch size among the enclosures, ranging from 5.5 - 10 eggs per clutch for *D. leptopus* and from 14 - 52 eggs per clutch for *D. kenai*. But even more convincing is the fact that, by the second year, the enclosures had established *D. kenai* populations of very different size structures (with mean female cephalothorax length ranging from 0.98 mm in the No Fertilizer enclosure to 1.25 mm in the 10X enclosure). In addition, during the second year there were no longer differences among the enclosures in lipid stores, suggesting that the phytoplankton communities present in each enclosure were equally sufficient in supporting energy and nutritional demands of the resident copepod populations. These results suggest that the two size classes differ in their physiological sensitivity to food quality.

While the evidence is highly suggestive that morphological, behavioral, and physiological differences between the two subpopulations of *D. kenai* may enhance persistence of the two size classes in nature, it is possible that some other factor not studied here may also

contribute to the coexistence and persistence of the two size classes, the most obvious being predation. Predation is a strong driving force in the aquatic environment, inducing morphological variation in the prey (Gilbert 1966; Krueger and Dodson 1981; Dodson 1989), predator-avoiding vertical migration patterns (Haney 1988; Neill 1990), or changes in the size structure of the prey populations (Brooks and Dodson 1965; Vanni 1987; Vanni and Findlay 1990). Northcote *et al.* (1978) reported a decrease in the mean body size of *D. kenai* and in *Chaoborus* population density in U.B.C. Research Forest lakes to which fish had been added. While size selective predation would seem to be the most obvious explanation for the change in mean body size, their evidence did not support that theory; fish gut contents provided no evidence of size selection and the small body size persisted even "after fish predation became negligible" (Northcote *et al.* 1978). They suggested, but did not test, the alternative hypothesis that the change in size was evidence of competition-induced decreased growth rate, as a consequence of increased population density under decreasing *Chaoborus* predation pressure.

Combining the conclusions of Northcote *et al.* (1978) with the results of my research suggests another possible mechanism responsible for maintaining two size classes in one case and eliminating one size class in another. The fish added to the lakes in the study by Northcote *et al.* substantially reduced, and subsequently eliminated, the *Chaoborus* populations from the lakes. The two species which occur in the lakes, *C. americanus* and *C. trivittatus*, are themselves effective predators upon freshwater zooplankton and are distinguished by two characteristics. *C. trivittatus* is larger in size and has a longer life cycle (two year cycle versus 1 year cycle) than the smaller *C. americanus* (Fedorenko and Swift 1972). As a consequence of the difference in size, the two species differ in predatory behavior. Of particular interest for the purpose of this argument is the difference in predation on *D. kenai*; the larger *C. trivittatus* can effectively ingest *D. kenai* upon reaching the third larval instar, while the smaller *C. americanus* can ingest *D. kenai* only when it has reached the fourth instar (Fedorenko 1975a,b; Swift and Fedorenko 1975). Unfortunately, these studies did not recognize any variation in size within

the *D. kenai* population nor did they mention whether variation in *D. kenai* size affected susceptibility to predation. However, it seems quite likely that, if the difference in mouth gape of 0.04 mm which distinguishes third instar *C. trivittatus* and *C. americanus* (Swift and Fedorenko 1975) is sufficient to affect predation on *D. kenai*, then differences between the two sizes of *D. kenai* might be sufficient to affect relative risk of predation by the two *Chaoborus* species. There is suggestive evidence from the study of Northcote *et al.* (1978) that such a difference might exist. When fish were added to the lakes, *C. americanus*, which tended to remain high in the water column, was eliminated during the first year whereas the larger *C. trivittatus*, which tended to remain deeper in the water column, was not eliminated until the second year. However, the difference in mean body size was established during the first year, while *C. trivittatus* was still present in the lakes, indicating the small *D. kenai*, but not the large, were able to persist in the presence of the larger *Chaoborus*. Thus, with removal of the smaller *Chaoborus*, the small *D. kenai* was released from the controlling effects of predation while predation on large *D. kenai* remained unaffected.

Regardless of the ecological processes which may favor one morphotype over another, the temporal patterns of reproduction separate the two size groups into what could be considered distinct species, according to the biological species concept (see reviews by Scudder 1974 and Templeton 1989). Although there is some overlap in the late summer, for the most part the two groups are reproductively isolated over time. Females produce offspring which are similar in size to the mother, suggesting that females have mated with similarly sized males to produce similarly sized offspring, again suggesting that the two groups are reproductively isolated. There is no detectable difference in egg size between the two size groups (i.e., smaller females do not produce smaller eggs); therefore it does not appear that the difference in offspring size is a consequence of starting off as a smaller nauplius. Similar results have been reported for a population of isopod, *Asellus* (Thompson 1986). Non-random mating, coupled with heritability of body size, resulted in a positive genetic feedback mechanism which promoted wide variation in size-frequency distribution. While I have not

tested for genetic differences between the two groups, it seems possible that the two groups may well be undergoing sympatric speciation from the original *Diaptomus kenai* population. Although the concept of sympatric speciation is controversial (see review of Tauber and Tauber 1989), it is recognized as a distinct process of speciation (Tauber and Tauber 1981; Snell and Hawkins 1983; Rice and Salt 1988).

In conclusion, it appears that the coexistence of the two size classes is due to the interactions of two processes. The results presented here suggest that the two sizes differ in their utilization of available food resources, given the subtle differences in feeding behavior. Possibly, as a consequence of this difference, they differ in their perception of the quality of the available food, as evidenced by the variation among the enclosures in lipid stores during the first year (when both size classes were present) and the variation among the enclosures in size class composition in the second year. In addition, by virtue of their size, they differ in susceptibility to predation by *Chaoborus*, as interpreted from the results of Northcote *et al.* (1978). While this is the first study to report on the existence of reproductively distinct, co-occurring size classes within a single copepod species, I suspect this is not a unique phenomenon, but rather an overlooked one. Data from other oligotrophic lakes in the Research Forest (e.g., M.A. Chapman, Univ. Waikato, unpubl. data; C.J. Walters, U.B.C., unpubl. data) suggest that bimodal size distributions may exist in other populations of *D. kenai*. Whether this is an attribute unique to *D. kenai* or unique to populations in oligotrophic systems is a question worthy of further investigation.

These results demonstrate that *D. kenai* and *D. leptopus* are extremely flexible and adaptable animals. They modify their behavior, morphology, and physiology over a range of magnitudes and time scales, bringing into question the utility of models generated under laboratory conditions in predicting behaviors or dynamics of copepod populations or communities in nature. There can be very subtle changes in feeding behavior (such as reported in Chapter Four) or very pronounced changes in size structure (as described in

Chapter Two). The time scales involved can vary from hours (e.g., the changes in filtering rates and electivities reported in Chapter Four) to days (e.g., the changes in clutch size reported in Chapter Two or changes in lipid levels reported in Chapter Three) or seasons (e.g., the shift in *D. kenai* population size structure reported in Chapter Two). As desirable as it may be to "compartmentalize nature" according to the simple limitation model of Williamson and Butler (1987), or the size-structured feeding processes presented by Vanderploeg (1981) and Vanderploeg *et al.* (1984), it is clear that there is a complex interaction of factors which influence the physiology, morphology, and behavior of copepods in nature.

LITERATURE CITED

Abou Debs, C. 1984. Carbon and nitrogen budget of the calanoid copepod *Temora stylifera*; effect of concentration and composition of food. Mar. Ecol. Progr. Ser. 15: 213-223.

Abou Debs, C. and P. Nival. 1983. Étude de la ponte et du développement embryonnaire en relation avec la température et la nourriture chez *Temora stylifera* Dana (Copepoda: Calanoida). J. Exp. Mar. Biol. Ecol. 72: 125-145.

Abrams, P.A. 1990. The effects of adaptive behavior on the type-2 functional response. Ecology 71: 877-885.

Admiraal, W. and D. Werner. 1983. Utilization of limiting concentrations of ortho-phosphate and production of extracellular organic phosphates in cultures of marine diatoms. J. Plank. Res. 5: 495-513.

Admiraal, W., H. Peletier, and R.W.P.M. Laane. 1986. Nitrogen metabolism of marine planktonic diatoms; excretion assimilation and cellular pools of free amino acids in seven species with different cell size. J. Exp. Mar. Biol. Ecol. 98: 241-263.

Alcaraz, M., G.-A. Paffenhöfer, and J.R. Strickler. 1980. Catching the algae: a first account of visual observations on filter-feeding calanoids. ASLO Spec. Symp. 3: 241-248.

Allan, J.D. and C.E. Goulden. 1980. Some aspects of reproductive variation among freshwater zooplankton. ASLO Spec. Symp. 3: 388-410.

Anderson, R.S. 1967. Diaptomid copepods from two mountain ponds in Alberta. Can. J. Zool. 45: 1043-1047.

Anderson, R.S. 1970. Predator-prey relationships and predation rates for crustacean zooplankters from some lakes in Canada. Can. J. Zool. 48: 1229-1240.

Andrews, J.C. 1983. Deformation of the active space in the low Reynolds number feeding current of calanoid copepods. Can. J. Fish. Aquat. Sci. 40: 1293-1302.

Armitage, K.B., B. Saxena, and E.E. Angino. 1973. Population dynamics of pond zooplankton, I. *Diaptomus pallidus* Herrick. Hydrobiologia 42: 295-333.

Arnold, D.E. 1971. Ingestion, assimilation, survival, and reproduction by *Daphnia pulex* fed seven species of blue-green algae. Limnol. Oceanogr. 16: 906-920.

Arnott, G.H., G.W. Brand, and L.C. Kos. 1986. Effects of food quality and quantity on the survival, development and egg production of *Gladioferens pectinatus* (Copepoda: Calanoida). Australian J. Mar. Freshw. Res. 37: 467-474.

Arts, M.T. and W.G. Sprules. 1987. Energy reserves of three zooplankton species from two lakes with different metal concentrations. Can. J. Fish. Aquat. Sci. 44: 458-466.

Arts, M.T. and W.G. Sprules. 1988. Evidence for indirect effects of fish predation on maternal lipid investment in *Holopedium gibberum*. Can. J. Fish. Aquat. Sci. 45: 2147-2155.

Barica, J., H. Kling, and J. GIbson. 1980. Experimental manipulation of algal bloom composition by nitrogen addition. Can. J. Fish. Aquat. Sci. 37: 1175-1183.

Ben-Amotz, A., R. Fishler, and A. Schneller. 1987. Chemical composition of dietary species

of marine unicellular algae and rotifers with emphasis on fatty acids. Mar. Biol. 95: 31-36.

Ben-Amotz, A., T.G. Tornabene, and W.H. Thomas. 1985. Chemical profile of selected species of microalgae with emphasis on lipids. J. Phycol. 21: 72-81.

Benson, A.A. and R.F. Lee. 1975. The role of wax in oceanic food chains. Sci. Amer. 232: 76-86.

Benson, A.A., R.F. Lee, and J.C. Nevenzel. 1972. Wax esters: major marine metabolic energy sources. *In*: J. Ganguly and R.M.S. Smellic (eds.) Current Trends in the Biochemistry of Lipids. Academic Press, London. pp. 175-187.

Bergquist, A.M., S.R. Carpenter, and J.C. Latino. 1985. Shifts in phytoplankton size structure and community composition during grazing by contrasting zooplankton assemblages. Limnol. Oceanogr. 30: 1037-1045.

Björnberg, T.K.S. 1986. Aspects of the appendages in development. Syllogeus 58: 51-66.

Black, R.W.II and N.G. Hairston, Jr. 1988. Predator driven changes in community structure. Oecologia 77: 468-479.

Blades-Eckelberger, P.I. 1986. Aspects of internal anatomy and reproduction in the Copepoda. Syllogeus 58: 26-50.

Boyd, C.M. 1976. Selection of particle sizes by filter-feeding copepods: A plea for reason. Limnol. Oceanogr. 21: 175-180.

Brendelberger, H. 1985. Filter mesh-size and retention efficiency for small particles: comparative studies with Cladocera. Arch. Hydrobiol. Beih. 21: 135-146.

Brooks, J.L. and S.I. Dodson. 1965. Predation, body size, and composition of plankton. Science 150: 28-35.

Buckingham, S.L. 1978. Functional responses and feeding strategies of fresh-water filter-feeding zooplankton. Dissertation, University of British Columbia, 158pp.

Butler, N.M., C.A. Suttle, and W.E. Neill. 1989. Discrimination by freshwater zooplankton between single algal cells differing in nutritional status. Oecologia 78: 368-372.

Cahoon, L.B. 1981. Reproductive response of *Acartia tonsa* to variations in food ration and quality. Deep Sea Res. 28A: 1215-1221.

Carefoot, T.H. 1984a. Nutrition and growth of *Ligia pallasii*. Symp. Zool. Soc. Lond. 53: 455-467.

Carefoot, T.H. 1984b. Studies on the nutrition of the supralittoral isopod *Ligia pallasii* using chemically defined artificial diets: assessment of vitamin, carbohydrate, fatty acid, cholesterol, and mineral requirements. Comp. Biochem. Physiol. 79A: 655-665.

Cargill, A.S. II, K.W. Cummins, B.J. Hanson, and R.R. Lowry. 1985. The role of lipids as feeding stimulants for shredding aquatic insects. Freshwater Biol. 15: 455-464.

Carpenter, S.R., J.F. Kitchell, J.R. Hodgson, P.A. Cochran, J.J. Elser, M.M. Elser, D.M. Lodge, D. Kretchmer, X. He, and C.N. vonEnde. 1987. Regulation of lake primary productivity by food web structure. Ecology 68: 1863-1876.

Cavaletto, J.F., H.A. Vanderploeg, and W.S. Gardner. 1989. Wax esters in two species of freshwater zooplankton. Limnol. Oceanogr. 34: 785-789.

Chapman, D. 1969. Introduction to Lipids. McGraw-Hill, London.

Charnov, E.L. and G.H. Orians. 1973. Optimal foraging: Some theoretical explorations. Univ. Washington, Seattle. 160pp.

Checkley, D.M. Jr. 1980. Food limitation of egg production by a marine planktonic copepod in the sea off southern California. Limnol. Oceanogr. 25: 991-998.

Chow-Fraser, P. 1986. An empirical model to predict in situ grazing rates of *Diaptomus minutus* Lilljeborg on small algal particles. Can. J. Fish. Aquat. Sci. 43: 1065-1070.

Chow-Fraser, P. and C.K. Wong. 1985. Herbivorous feeding of three large freshwater calanoid copepods, *Limnocalanus macrurus* Sars, *Senecella calanoides* Juday, and *Epischura lacustris* Forbes. Verh. Internat. Verein. Limnol. 22: 3195-3198.

Chu, F.E. and K.L. Webb. 1984. Polyunsaturated fatty acids and neutral lipids in developing larvae of the oyster, *Crassostrea virginica*. Lipids 19: 815-820.

Clarke, A., A. Skadsheim, and L.J. Holmes. 1985. Lipid biochemistry and repoductive biology in two species of Gammaridae (Crustacea: Amphipoda). Mar. Biol. 88: 247-263.

Comita, G.W. 1964. The energy budget of *Diaptomus siciloides*, Lilljeborg. Verh. Internat. Verein. Limnol. 15: 646-653.

Conklin, E.D. and L. Provasoli. 1977. Nutritional requirements of the water flea *Moina macrocopa*. Biol. Bull. 152: 337-350.

Conover, R.J. 1962. Metabolism and growth in *Calanus hyperboreus* in relation to its life cycle. Rapp. Proc.-Verb. Cons. Int. Explor. Mer. 153: 190-197.

Conover, R.J. 1968. Zooplankton - life in a nutritionally dilute environment. Amer. Zool. 8: 107-118.

Conover, R.J. 1978. Transformation of organic matter. *In*: O. Kinne (ed.) Marine Ecology IV. Dynamics. J. Wiley and Sons, Chichester. pp. 221-499.

Corkett, C.J. and I.A. McLaren. 1978. The bioloty of *Pseudocalanus*. Adv. Mar. Biol. 15: 1-231.

Cowgill, U.M., D.M. Williams, and J.B. Esquivel. 1984. Effects of maternal nutrition on fat content and longevity of neonates of *Daphnia magna*. J. Crust. Biol. 4: 173-190.

Cowgill, U.M., D.L. Hopkins, S.L. Applegath, I.T. Takahashi, S.D. Brooks, and D.P. Milazzo. 1985a. Brood size and neonate weight of *Daphnia magna* produced by nine diets. *In*: R.C. Bahner and D.J. Hansen (eds.) Aquatic Toxicology and Hazard Assessment: Eighth Symposium. Amer. Soc. Test. Mat., Phila. pp. 233-244.

Cowgill, U.M., K.I. Keating, and I.T. Takahashi. 1985b. Fecundity and longevity of *Ceriodaphnia dubia/affinis* in relation to diet at two different temperatures. J. Crust. Biol. 5: 420-429.

Cowles, T.J. and J.R. Strickler. 1983. Characterization of feeding activity patterns in the

planktonic copepod *Centropages typicus* Kroyer under various food conditions. Limnol. Oceanogr. 28: 106-115.

Cowles, T.J., R.J. Olson, and S.W. Chisholm. 1988. Food selection by copepods: discrimination on the basis of food quality. Mar. Biol. 100: 41-49.

Crawford, P.and G.R. Daborn. 1986. Seasonal variations in body size and fecundity in a copepod of turbid estuaries. Estuaries 9: 133-141.

Crowder, L.B., M.E. McDonald, and J.A. Rice. 1987. Understanding recruitment of Lake Michigan fishes: the importance of size-based interactions between fish and zooplankton. Can. J. Fish. Aquat. Sci. 44 (Suppl. 2): 141-147.

Crumpton, W.G. and R.G. Wetzel. 1982. Effects of differential growth and mortality in the seasonal succession of phytoplankton populations. Ecology 63: 1729-1739.

Culver, D. 1980. Seasonal variation in the sizes at birth and at first reproduction in Cladocera. ASLO Spec. Symp. 3: 358-366.

D'Abramo, L.R. 1979. Dietary fatty acid and temperature effects of the productivity of the cladoceran, *Moina macrocopa*. Biol. Bull. 157: 234-248.

Dagg, M.J. 1985. The effects of food limitation on diel migratory behavior in marine zooplankton. Arch. Hydrobiol. Beih. 21: 247-255.

Dagg, M.J. and W.E. Walser, Jr. 1987. Ingestion, gut passage, and egestion by the copepod *Neocalanus plumchrus* in the laboratory and in the subarctic Pacific Ocean. Limnol. Oceanogr. 32: 178-188.

Daro, M.H. 1985. Feeding rhythms and vertical distribution of marine copepods. Bull. Mar. Sci. 37: 487-497.

Davis, C.C. 1984. Planktonic Copepoda (Including Monstrilloida). *In*: K.A. Steidinger and L.M. Walker (eds.) Marine plankton Life Cycle Strategies. C.R.C. Press, pp. 67-91.

DeMott, W.R. 1986. The role of taste in food selection by freshwater zooplankton. Oecologia (Berl.) 69: 334-340.

DeMott, W.R. 1988a. Discrimination between algae and artificial particles by freshwater and marine copepods. Limnol. Oceanogr. 33: 397-408.

DeMott, W.R. 1988b. Discrimination between algae and detritus by freshwater and marine copepods. Bull. Mar. Sci. 43: 486-499.

DeMott, W.R. 1989. Optimal foraging theory as a predictor of chemically mediated food selection by suspension-feeding copepods. Limnol. Oceanogr. 34: 130-154.

Dickman, M. and I.E. Efford. 1972. Some effects of artificial fertilization on enclosed plankton populations in Marion Lake, British Columbia. Can. J. Fish. Aquat. Sci. 29: 1595-1604.

Digby, P.G.N. and R.A. Kempton. 1987. Multivariate Analysis of Ecological Communities. Chapman and Hall, NYC. 206pp.

Dodson, S. 1989. Predator-induced reaction norms. BioScience 39: 447-452.

Donaghay, P.L. 1988. Role of temporal scales of acclimation, food quality, and trophic dominance in controlling the evolution of copepod feeding behavior. Bull. Mar. Sci. 43: 469-485.

Dortch, Q. 1987. The biochemical composition of plankton in a subsurface chlorophyll maximum. Deep-Sea Res. 34: 705-712.

Durbin, E.G., A.G. Durbin, T.J. Smayda, and P.G. Verity. 1983. Food limitation of production by adult *Acartia tonsa* in Narragansett Bay, Rhode Island. Limnol. Oceanogr. 28: 1199-1213.

Edmondson, W.T. 1959. Freshwater Biology. J. Wiley and Sons, NY. 1248pp.

Edmondson, W.T., G.W. Comita, and G.C. Anderson. 1962. Reproductive rate of copepods in nature and its relation to phytoplankton population. Ecology 43: 625-634.

Elmore, J.L. 1982. The influence of food concentration and container volume on life history parameters of *Diaptomus dorsalis* from subtropical Florida. Hydrobiologia 89: 215-223.

Elrifi, I.R. and D.H. Turpin. 1985. Steady-state luxury consumption and the concept of optimum nutrient ratios: a study with phosphate and nitrate limited *Selenastrum minutum* (Chlorophyta). J. Phycol. 21: 592-602.

Engen, S. and N.C. Stenseth. 1984. An ecological paradox: A food type may become more rare in the diet as a consequence of being more abundant. Am. Nat. 124: 352-359.

Evans, F. 1981. An investigation into the relationship of sea temperature and food supply to the size of planktonic copepod *Temora longicornis* Müller in the North Sea. Est. Coast. Shelf Sci. 13: 145-158.

Evans, M.S. 1986. Recent major declines in zooplankton populations in the inshore region of Lake Michigan: probable causes and implications. Can. J. Fish. Aquat. Sci. 43: 154-159.

Evans, M.S., G.J. Warren, and J. Glick. 1986. Population dynamics of copepod zooplankton in southeastern Lake Michigan, with considerations of overwintering strategies. Syllogeus 58: 629.

Falk-Petersen, S., R.R. Gatten, J.R. Sargent, and C.C.E. Hopkins. 1981. Ecological investigations on the zooplankton community in Balsfjorden, Northern Norway: Seasonal changes in the lipid class composition of *Meganyctiphanes norvegica* (M.Sars), *Thysanoessa raschii* (M. Sars), and *T. inermis* (Kroyer). J. Exp. Mar. Biol. Ecol. 54: 209-224.

Fedorenko, A.Y. 1975a. Feeding characteristics and predation impact of *Chaoborus* (Diptera, Chaoboridae) larvae in a small lake. Limnol. Oceanogr. 20: 250-258.

Fedorenko, A.Y. 1975b. Instar and species-specific diets in two species of *Chaoborus*. Limnol. Oceanogr. 20: 238-249.

Fedorenko, A.Y. and M.C. Swift. 1972. Comparative biology of *Chaoborus americanus* and *Chaoborus trivittatus* in Eunice Lake, British Columbia. Limnol. Oceanogr. 17: 721-730.

Fogg, G.E. 1959. Nitrogen nutrition and metabolic patterns in algae. Symp. Soc. Exp. Biol. 13: 106-125.

Frost, B.W. 1972. Effects of size and concentration of food particles on the feeding behavior of the marine planktonic copepod *Calanus pacificus*. Limnol. Oceanogr. 17: 805-815.

Frost, B.W. 1977. Feeding behavior of *Calanus pacificus* in mixtures of food particles. Limnol. Oceanogr. 22: 472-491.

Frost, B.W. 1985. Food limitation of the planktonic marine copepods *Calanus pacificus* and *Pseudocalanus* sp. in a temperate fjord. Arch. Hydrobiol. Beih. 21: 1-13.

Fulton, R.S. III and H.W. Paerl. 1987. Toxic and inhibitory effects of the blue-green alga *Microcystis aeruginosa* on herbivorous zooplankton. J. Plank. Res. 9: 837-855.

Fulton, R.S. III and H.W. Paerl. 1988. Zooplankton feeding selectivity for unicellular and colonial *Microcystis aeruginosa*. Bull. Mar. Sci. 43: 500-508.

Furnadzieva, S., G.D. Petkov, and S.S. Popov. 1987. Lipids of *Scenedesmus incrassatulus* R-83 cultivated under various technological regimes. Bulgarian Acad. Sci. Hydrob. 29: 3-12.

Gallager, S.M. and R. Mann. 1986. Individual variability in lipid content of bivalve larvae quantified histochemically by absorption photometry. J. Plankton Res. 8: 927-937.

Gardner, W.S., W.A. Frez, E.A. Cichocki, and C.C. Parrish. 1985a. Micromethod for lipids in aquatic invertebrates. Limnol. Oceanogr. 30: 1099-1105.

Gaudy, R. et J.P. Guérin. 1977. Dynamique des populations de *Tisbe holothuriae* (Crustacea: Copepoda) en élevage sur trois régimes artificiels différents. Mar. Biol. 39: 137-145.

Gehrs, C.W. and A. Robertson. 1975. Use of life tables in analyzing the dynamics of copepod populations. Ecology 56: 665-672.

Gilbert, J.J. 1966. Rotifer ecology and embryological induction. Science 151: 1234-1237.

Gilbert, L.I. and J.D. O'Connor. 1970. Lipid metabolism and transport in arthropods. *In*: M. Florkin and B.T. Scheer (eds.) Chemical Zoology, V.5, Arthropoda Part A. Academic Press, NY.

Gliwicz, Z.M. and W. Lampert. 1990. Food thresholds in *Daphnia* species in the absence and presence of blue-green filaments. Ecology 71: 691-702.

Goad, L.J. 1976. The steriods of marine algae and invertebrate animals. *In*: D.C. Malins and J.R. Sargent (eds.) Biochemical and Biophysical Perspectives in Marine Biology. Academic Press, NY.

Goulden, C.E. and L. Henry. 1984. Lipid energy reserves and their role in Cladocera. AAAS Sel. Symp. 85: 167-185.

Goulden, C.E., L. Henry, and A.J. Tessier. 1982. Body size, energy reserves, and competitive ability in three species of Cladocera. Ecology 63: 1780-1789.

Grigg, H., S.J. Bardwell, and L.J. Holmes. 1987. Comparative observations on the biometry and development of *Calanus finmarchicus* and *C. helgolandicus* in Copepodite Stage V, with comments on other Calanidae. Mar. Biol. 96: 253-262.

Hadley, N.F. 1981. Cuticular lipids of terrestrial plants and arthropods: A comparison of their structure, composition, and waterproofing function. Biol. Rev. 56: 23-47.

Hadley, N.F. 1985. The Adaptive Role of Lipid in Biological Systems. J. Wiley and Sons, NY.

Hagen, W. 1988. Zur Bedeutung der Lipide im antarktischen Zooplankton. Ber. Polarforsch. 49: 1-129.

Håkanson, J.L. 1984. The long and short term feeding condition in field-caught *Calanus pacificus*, as determined from the lipid content. Limnol. Oceanogr. 29: 794-804.

Håkanson, J.L. 1987. The feeding condition of *Calanus pacificus* and other zooplankton in relation to phytoplankton pigments in the California Current. Limnol. Oceanogr. 32: 881-894.

Hall, D.J. 1964. An experimental approach to the dynamics of a natural population of *Daphnia galeata mendotae*. Ecology 45: 94-112.

Hammer, V.T. and W.W. Sawchyn. 1968. Seasonal succession and congenic associations of *Diaptomus* spp. in Saskatchewan ponds. Limnol. Oceanogr. 13: 476-484.

Haney, J.F. 1988. Diel patterns of zooplankton behavior. Bull. Mar. Sci. 43: 583-603.

Haney, J.F. and M.A. Trout. 1985. Size selective grazing by zooplankton in Lake Titicaca. Arch. Hydrobiol. Beih. 21: 147-160.

Hanson, B.J., K.W. Cummins, A.S. Cargill, and R.R. Lowry. 1983. Dietary effects on lipid and fatty acid composition of *Clistoronia magnifica* (Trichoptera: Limniphilidae). Freshwater Invert. Biol. 2: 2-15.

Harrison, P.J., H.L. Conway, R.W. Holmes, and C.O. Davis. 1977. Marine diatoms grown in chemostats under silicate or ammonium limitation. III. Cellular chemical composition and morphology of *Chaetoceros debilis*, *Skeletonema costatum*, and *Thalassiosira gravida*. Mar. Biol. 43: 19-31.

Hartmann, H.J. 1985. Feeding of *Daphnia pulicaria* and *Diaptomus ashlandi* on mixtures of unicellular and filamentous algae. Verh. Internat. Verein. Limnol. 22: 3178-3183.

Hawkins, P. and W. Lampert. 1989. The effect of *Daphnia* body size on filtering rate inhibition in the presence of a filamentous cyanobacterium. Limnol. Oceanogr. 34: 1084-1089.

Head, E.J.H. and L.R. Harris. 1985. Physiological and biochemical changes in *Calanus hyperboreas* from Jones Sound NWT during the transition from summer feeding to overwintering condition. Polar Biol. 4: 99-106.

Henderson, R.J. and J.R. Sargent. 1980. Biosynthesis of neutral lipids by *Euchaeta norvegica*. Mar. Bio. 56: 1-6.

Herzig, A., R.S. Anderson, and D.W. Mayhood. 1980. Production and population dynamics of *Leptodiaptomus sicilis* in a mountain lake in Alberta, Canada. Holarct. Ecol. 3: 50-63.

Hoenicke, R. and C.R. Goldman. 1987. Resource dynamics and seasonal changes in competitive interactions among three cladoceran species. J. Plank. Res. 9: 397-417.

Holling, C.S. 1959. The components of predation as revealed by a study of small-mammal predation of the European pine sawfly. Can. Ent. 91: 293-320.

Holling, C.S. 1966. The functional response of invertebrate predators to prey density. Mem. Ent. Soc. Can. 48: 1-86.

Holm, N.P. and J. Shapiro. 1984. An examination of lipid reserves and the nutritional status of *Daphnia pulex* fed *Aphanizomenon flos-aquae*. Limnol. Oceanogr. 29: 1137-1140.

Houde, S.E.L. and M.R. Roman. 1987. Effects of food quality on the functional ingestion response of the copepod *Acartia tonsa*. Mar. Ecol. Prog. Ser. 40: 69-77.

Huntley, M. 1985. A method for estimating food-limitation and potential production of zooplankton communities. Arch. Hydrobiol. Beih. 21: 41-55.

Huntley, M.E., P. Ciminiello, and M.D.G. Lopez. 1987. Importance of food quality in determining development and survival of *Calanus pacificus* (Copepoda: Calanoida). Mar. Biol. 95: 103-113.

Hutchinson, G.E. 1967. A Treatise on Limnology. Vol II. J. Wiley and Sons, NY.

Ikeda, T. 1974. Nutritional ecology of marine zooplankton. Mem. Fac. Fish. Hokkaido Univ. 22: 1-97.

Istvánovics, V., L. Vörös, S. Herodek, L.G. Tóth, and I. Tátrai. 1986. Changes of phosphorous and nitrogen concentration and of phytoplankton in enriched lake enclosures. Limnol. Oceanogr. 31: 798-811.

Ivlev, V.S. 1961. Experimental Ecology of the Feeding of Fishes. Yale Univ. Press, New Haven.

Jacobs, J. 1977a. Coexistence of similar zooplankton species by differential adaptation to reproduction and escape in an environment with fluctuating food and enemy densities. I. A model. Oecologia (Berl.) 29: 233-247.

Jacobs, J. 1977b. Coexistence of similar zooplankton species by differential adaptation to reproduction and escape in an environment with fluctuating food and enemy densities. II. Field data analysis of *Daphnia*. Oecologia (Berl.) 29: 233-247.

Jespersen, D. 1924. On the frequency of birds over the high Atlantic Ocean. Nature 114: 281-283.

Johnsen, G.H. and P.J. Jakobsen. 1987. The effect of food limitation on vertical migration in *Daphnia longispina*. Limnol. Oceanogr. 32: 873-880.

Johnson, J.K. 1980. Effects of temperature and salinity on production and hatching of dormant eggs of *Acartia californiensis* (Copepoda) in an Oregon estuary. Fish. Bull. 77: 567-584.

Jones, K.J., P. Tett, A.C. Wallis, and B.J. Wood. 1978. Investigations of a nutrient-growth model using a continuous culture of natural phytoplankton. J. Mar. Biol. Assoc. U.K. 58: 923-941.

Jones, D.A., A. Kanazawa, and K. Ono. 1979. Studies on the nutritional requirements of the larval stages of *Penaeus japonicus* using microencapsulated diets. Mar. Biol. 54: 261-267.

Kerfoot, W.C. 1974. Net accumulation rates and the history of cladoceran communities. Ecology 55: 51-61.

Kerfoot, W.C. and W.R. DeMott. 1980. Foundations for evaluating community interactions: the use of enclosures to investigate the coexistence of *Daphnia* and *Bosmina*. ASLO Spec. Symp. 3: 725-741.

Kilham, S.S. 1986. Dynamics of Lake Michigan natural phytoplankton communities in continuous cultures along a Si:P loading gradient. Can. J. Fish. Aquat. Sci. 43: 351-360.

Kilham, S.S. and P. Kilham. 1984. The importance of resource supply rates in determing phytoplankton community structure. AAAS Sel. Symp. 85: 7-27.

Kimmerer, W.J. 1984. Spatial and temporal variability in egg production rates of the calanoid copepod *Acrocalanus inermis*. Mar. Biol. 78: 165-169.

Kimmerer, W.J. and A.D. McKinnon. 1987. Growth, mortality, and secondary production of the copepod *Acartia tranteri* in Westernport Bay, Australia. Limnol. Oceanogr. 32: 14-28.

Klein Breteler, W.C.M. and S.R. Gonzalez. 1988. Influence of temperature and food concentration on body size, weight and lipid content of two Calanoid copepod species. Hydrobiologia 167/168: 201-210.

Klinka, K. 1976. Ecosystem map of the University of British Columbia Research Forest. Faculty of Forestry, U.B.C., Vancouver.

Knoechel, R. and J. Kalff 1978. An in situ study of the species productivity and population dynamics of five planktonic diatoms. Limnol. Oceanogr. 23: 195-218.

Koehl, M.A.R. and J.R. Strickler. 1981. Copepod feeding currents: food capture at low Reynolds number. Limnol. Oceanogr. 26: 1062-1073.

Kogeler, J.W., S. Falk-Petersen, A. Kristensen, F. Pettersen, and J. Dahlen. 1987. Density and sound speed contrasts in sub-arctic zooplankton. Polar Biol. 7: 231-235.

Kondratieff, P. and G.M. Simmons. 1984. Nutritive quality and size fractions of natural seston in an impounded river. Arch. Hydrobiol. 101: 401-412.

Krebs, C.J. 1989. Ecological Methodology. Harper and Row, NY. 654pp.

Krebs, J.R., D.W. Stephens, and W.J. Sutherland. 1983. Perspectives in optimal foraging. *In*: A.H. Brush and G.A. Clark Jr. (eds.) Perspectives in Ornithology. Cambridge Univ. Press, Cambridge. pp.165-216.

Krueger, D.A. and S.I. Dodson. 1981. Embryological induction and predation ecology in *Daphnia pulex*. Limnol. Oceanogr. 26: 219-223.

Laing, I. and P.F. Millican. 1986. Relative growth and growth efficiency of *Ostrea edulis* spat fed various algal diets. Aquaculture 54: 245-262.

Lampert, W. 1977. Studies on the carbon balance of *Daphnia pulex* de Geer as related to environmental conditions. IV. Determination of the "threshold" concentration as a factor controlling the abundance of zooplankton species. Arch. Hydrobiol. Suppl. 48: 361-368.

Lampert, W. 1981. Inhibitory and toxic effects of blue-green algae on *Daphnia*. Int. Rev. Ges. Hydrobiol. 66: 285-298.

Lampert, W. 1987. Feeding and nutrition in *Daphnia*. Mem. Ist. Ital. Idrobiol. 45: 143-192.

Lampert, W. 1988. The relative importance of food limitation and predation in the seasonal cycle of two *Daphnia* species. Verh. Internat. Verein. Limnol. 23: 713-718.

Lampert, W. and U. Schober. 1980. The importance of "threshold" food concentrations. ASLO Spec. Symp. 3: 264-267.

Landry, M.R. 1981. Switching between herbivory and carnivory by the planktonic marine copepod *Calanus pacificus*. Mar. Biol. 65: 77-82.

Landry, M.R. and R.P. Hassett. 1985. Time scales in behavioral, biochemical, and energetic adaptations to food-limiting conditions by a marine copepod. Arch. Hydrobiol. Beih. 21: 209-221.

Lane, J.L. and C.R. Goldman. 1984. Size-fractionation of natural phytoplankton communities in nutrient bioassay studies. Hydrobiologia 118: 219-223.

Langeland, A. and H. Reinertsen. 1982. Interactions between phytoplankton and zooplankton in a fertilized lake. Holarctic Ecology 5: 253-272.

Lawrence, J.M. 1976. Patterns of lipid storage in post-metamorphic marine invertebrates. Amer. Zool. 16: 747-762.

Lechowicz, M.J. 1982. The sampling characteristics of electivity indices. Oecologia (Berl.) 52: 22-30.

Lee, R.F. 1974. Lipid composition of the copepod *Calanus hyperboreas* from the Arctic Ocean. Changes with depth and season. Mar. Biol. 26: 313-318.

Lee, R.F. 1975. Lipids of Arctic zooplankton. Comp. Biochem. Physiol. 51B: 263-266.

Lee, R.F. and A.T. Barnes. 1975. Lipids in the mesopelagic copepod, *Gaussia princeps*. Wax ester utilization during starvation. Comp. Biochem. Physiol. 52B: 265-268.

Lee, R.F. and J. Hirota. 1973. Wax esters in tropical zooplankton and nekton and the geographic distribution of wax esters in copepods. Limnol. Oceanogr. 18: 227-239.

Lee, R.F., J. Hirota, and A.M. Barnett. 1971a. Distribution and importance of wax esters in marine copepods and other zooplankton. Deep-Sea Res. 18: 1147-1165.

Lee, R.F., J. Hirota, J.C. Nevenzel, R. Sauerheber, A.A. Benson, and A. Lewis. 1972. Lipids in the marine environment. Calif. Mar. Res. Comm., CalCOFI Rep. 16: 95-102.

Lee, R.F., J. Nevenzel, and A.G. Lewis. 1974. Lipid changes during the life cycle of the marine copepod, *Euchaeta japonica* Marukawa. Lipids 9: 891-899.

Lee, R.F., J. Nevenzel, and G.-A. Paffenhöfer. 1971b. Importance of wax esters and other lipids in the marine food chain: phytoplankton and copepods. Mar. Biol. 9: 99-108.

Lee, W.Y., X.K. Zhang, C. VanBaalen, and C.R. Arnold. 1985. Feeding and reproductive performance of the harpacticoid *Tisbe carolinensis* (Copepoda, Crustacea) in four algal cultures. Mar. Ecol. Prog. Ser. 24: 273-279.

Legier-Visser, M.F., J.G. Mitchell, A. Okubo, and J.A. Fuhrman. 1986. Mechanoreception in calanoid copepods. A mechanism for prey detection. Mar. Biol. 90: 529-535.

Lehman, J.T. 1984. Grazing, nutrient release, and their impacts on the structure of phytoplankton communities. AAAS Sel. Symp. 85: 49-72.

Lehman, J.T. and C.D. Sandgren. 1985. Species-specific rates of growth and grazing loss among freshwater algae. Limnol. Oceanogr. 30: 34-46.

Lehman, J.T. and D. Scavia. 1982a. Microscale patchiness of nutrients in plankton communities. Science 216: 729-730.

Lehman, J.T. and D. Scavia. 1982b. Microscale nutrient patches produced by zooplankton. Proc. Natl. Acad. Sci. 79: 5001-5005.

Lehninger, A.L. 1975. Biochemistry. Worth Publishers, NY.

Lewis, R.W. 1970. The densities of three classes of marine lipids in relation to their possible role as hydrostatic agents. Lipids 5: 151-153.

Lynch, M., L.J. Weider, and W. Lampert. 1986. Measurement of the carbon balance in *Daphnia*. Limnol. Oceanogr. 31: 17-33.

MacArthur, R.H. and E.R. Pianka. 1966. On optimal use of a patchy environment. Am. Nat. 100: 603-609.

Mackie, P.R., K.J. Whittle, and R. Hardy. 1974. Hydrocarbons in the marine environment. I. *n*-Alkanes in the Firth of Clyde. Est. Coast. Mar. Sci. 2: 359-374.

Makarewicz, J.C. and G.E. Likens. 1975. Niche analysis of a zooplankton community. Science 190: 1000-1003.

Maly, E.J. 1973. Density, size, and clutch of two high altitude diaptomid copepods. Limnol. Oceanogr. 18: 840-848.

Maly, E.J. 1976. Resource overlap between co-occurring copepods: effects of predation and environmental fluctuation. Can. J. Zool. 54: 933-940.

Maly, E.J. 1983. Some further considerations on diaptomid body size and clutch size relationships. Limnol. Oceanogr. 28: 148-152.

Maly, E.J. and M.P. Maly. 1974. Dietary differences between two co-occurring calanoid copepod species. Oecologia 17: 325-333.

Marshall, S.M. and A.P. Orr. 1952. On the biology of *Calanus finmarchicus*. VII. Factors affecting egg production. J. Mar. Biol. Assoc. U.K. 30: 527-547.

Marshall, S.M. and A.P. Orr. 1955. The Biology of a Marine Copepod, *Calanus finmarchicus* (Gunnerus). Oliver and Boyd, Edinburgh.

Mayzaud, P. and S.A. Poulet. 1978. The importance of the time factor in the response of zooplankton to varying concentrations of naturally occurring particulate matter. Limnol. Oceanogr. 23: 1144-1154.

McLaren, I.A. 1965. Some relationships between temperature and egg size, body size, development rate, and fecundity, of the copepod *Pseudocalanus*. Limnol. Oceanogr. 10: 528-538.

McLaren, I.A., J.-M. Sevigny, and C.J. Corkett. 1988. Body sizes, development rates, and

genome sizes among *Calanus* species. Hydrobiologia 167/168: 275-284.

McQueen, D.J. 1970. Grazing rates and food selection in *Diaptomus oregonensis* (Copepoda) from Marion Lake, British Columbia. Can. J. Fish. Aquat. Sci. 27: 13-20.

Merrick, C.J. and G.G. Ganf. 1988. Effects of zooplankton grazing on phytoplankton communities in Mt. Bold Reservoir, South Australia, using enclosures. Australian J. Mar. Freshw. Res. 39: 503-523.

Meyer, A. 1987. Phenotypic plasticity and heterochrony in *Cichlasoma managuense* (Pisces, Cichlidae) and their implications for speciation in cichlid fishes. Evolution 41: 1357-1359.

Meyers, D.G. 1984. Habitat shifting, feeding mode versatility, and alternate resource exploitation by herbivorous cladoceran zooplankton in a montaine lake. AAAS Sel. Symp. 85: 309-345.

Moraitou-Apostolopoulou, M., G. Verriopoulous, and S. Hatjinikolaou. 1986. The population of planktonic copepods in two areas of Saronicos Gulf (Greece): population dynamics, contribution to secondary production, and relation to the principal oceanographic parameters. Syllogeus 58: 575-583.

Mullin, M.M., E.F. Stewart, and F.J. Fuglister. 1975. Ingestion by planktonic grazers as a function of the concentration of food. Limnol. Oceanogr. 20: 259-262.

Murdoch, W.W. 1969. Switching in general predators: Experiments on predator specificity and stability of prey populations. Ecol. Monogr. 39: 335-354.

Murdoch, W.W., S. Avery, and M.E.B. Smyth. 1975. Switching in predatory fish. Ecology 56: 1094-1105.

Myers, R.A. and J. Runge. 1986. Temperature-dependent changes in copepod adult size: an evolutionary theory. Syllogeus 58: 374-378.

Neill, W.E. 1978. Experimental studies on factors limiting colonization by *Daphnia pulex* Leydig of coastal montane lakes in British Columbia. Can. J. Zool. 56: 2498-2507.

Neill, W.E. 1981a. Developmental responses of juvenile *Daphnia rosea* to experimental alteration of temperature and natural seston concentration. Can. J. Fish. Aquat. Sci. 38: 1357-1362.

Neill, W.E. 1981b. Impact of *Chaoborus* predation upon the structure and dynamics of a crustacean zooplankton community. Oecologia (Berl.) 48: 164-177.

Neill, W.E. 1985. The effects of herbivore competition upon the dynamics of *Chaoborus* predation. Arch. Hydrobiol. Beih. 21: 483-491.

Neill, W.E. 1990. Induced vertical migration in copepods as a defense against invertebrate predation. Nature 345: 524-526.

Neill, W.E. and A. Peacock. 1980. Breaking the bottleneck: interactions of invertebrate predators and nutrients in oligotrophic lakes. ASLO Spec. Symp. 3: 715-725.

Nival, P. and S. Nival. 1973. Particle filtering efficiency of planktonic copepods. Ann. Inst. Oceanogr. 49: 135-144.

Nival, P. and S. Nival. 1976. Particle retention efficiencies of an herbivorous copepod, *Acartia clausi* (adult and copepodite stages): effects on grazing. Limnol. Oceanogr. 21: 24-38.

Nival, P. and S. Nival. 1979. Calculations of particle retention efficiency. Limnol. Oceanogr. 24: 995-998.

Nizan, S., C. Dimentman, and M. Shilo. 1986. Acute toxic effects of the cyanobacterium *Microcystis aeruginosa* on *Daphnia magna*. Limnol. Oceanogr. 31: 497-502.

Northcote, T.G., C.J. Walters, and J.M.B. Hume. 1978. Initial impacts of experimental fish introductions on the macrozooplankton of small oligotrophic lakes. Verh. Internat. Verein. Limnol. 20: 2003-2012.

Norusis, M.J. 1986. SPSS/PC+. SPSSinc. Chicago.

Norusis, M.J. 1988. SPSS/PC+ Advanced Statistics V2.0. SPSSinc. Chicago.

O'Connors, H.B. Jr., D.C. Biggs, and D.V. Minivaggi. 1980. Particle-size-dependent maximum grazing rates for *Temora longicornis* fed natural partical assemblages. Mar. Biol. 56: 65-70.

Ohman, M.D. 1988. Sources of variability in measurements of copepod lipids and gut fluorescence in the California Current coastal zone. Mar. Ecol. Prog. Ser. 42: 143-153.

O'Rourke, L. and R.E. Monroe. 1976. Utilization of radiolabelled acetate by three isopods in lipid synthesis. Comp. Biochem. Physiol. 55B: 553-557.

Paffenhöfer, G.-A. 1984. Calanoid copepod feeding: grazing on small and large particles. AAAS Sel. Symp. 85: 75-95.

Paffenhöfer, G.-A. 1988. Feeding rates and behavior of zooplankton. Bull. Mar. Sci. 43: 430-445.

Paffenhöfer, G.-A. and S.C. Knowles. 1978. Laboratory experiments on feeding, growth, and fecundity of, and effects of cadmium on *Pseudodiaptomus*. Bull. Mar. Sci. 28: 574-580.

Paffenhöfer, G.-A., J.R. Strickler, and M. Alcaraz. 1982. Suspension feeding by herbivorous calanoid copepods: A cinematographic study. Mar. Biol. 67: 193-199.

Parrish, C.C. 1987. Separation of aquatic lipid classes by Chromarod thin-layer chromatography with measurement by Iatroscan flame ionization detection. Can. J. Fish. Aquat. Sci. 44: 722-731.

Parrish, C.C. and R.G. Ackman. 1983a. Chromarod separations for the analysis of marine lipid classes by Iatroscan thin-layer chromatography-flame ionization detection. J. Chromatogr. 262: 103-112.

Parrish, C.C. and R.G. Ackman. 1983b. The effect of developing solvents on lipid class quantification in chromarod thin layer chromatography/flame ionization detection. Lipids 18: 563-565.

Parrish, C.C. and R.G. Ackman. 1985. Calibration of the Iatroscan-Chromarod system for marine lipid class analyses. Lipids 20: 521-530.

Parrish, C.C. and P.J. Wangersky. 1987. Particulate and dissolved lipid classes in cultures of *Phaeodactylum tricornutum* grown in cage culture turbidostats with a range of nitrogen supply rates. Mar. Ecol. Prog. Ser. 35: 119-128.

Parrish, C.C., X Zhou, and L.R. Herche. 1988. Flame ionization and flame thermionic detection of carbon and nitrogen in aquatic lipid and humic-type classes with an Iatroscan Mark IV. J. Chromatogr. 435: 350-356.

Patalas, J. and K. Patalas. 1978. Spatial variation in size and reproductive cycle of *Limnocalanus macrurus* in a deep, subarctic lake, Great Slave Lake. Verh. Internat. Verein. Limnol. 20: 150-158.

Pennak, R.W. 1978. Freshwater Invertebrates of the United States. J. Wiley and Sons, N.Y. 803pp.

Pessotti, E., C. Razouls, et S. Razouls. 1986. Distribution de taille d'une espéce de Copepode en relation avec sa distribution spatiale. Syllogeus 58: 409-419.

Peters, R.H. 1984. Methods for the study of feeding, grazing, and assimilation by zooplankton. *In*: J.A. Downing and F.H. Rigler (eds.) A Manual for the Assessment of Secondary Productivity in Fresh Waters. IBP Handbook 17, Blackwell Sci. Pub., Oxford.

Petersen, R. 1975. The paradox of the plankton: an equilibrium hypothesis. Am. Nat. 109: 35-49.

Peterson, W.T. 1985. Abundance, age structure, and in situ egg production rates of the copepod *Temora longicornis* in Long Island Sound, New York. Bull. Mar. Sci. 37: 726-738.

Pianka, E.R. 1976. Natural selection of optimal reproductive tactics. Amer. Zool. 16: 775-784.

Porter, K.G. and J.D. Orcutt, Jr. 1980. Nutritional adequacy, manageability, and toxicity as factors that determine the food quality of green and blue-green algae for *Daphnia*. ASLO Spec. Symp. 3: 268-281.

Porter, K.G., J. Gerritsen, and J.D. Orcutt, Jr. 1982. The effect of food concentration on swimming patterns, feeding behavior, ingestion, assimilation, and respiration by *Daphnia*. Limnol. Oceanogr. 27: 935-949.

Poulet, S.A. and P. Marsot. 1978. Chemosensory grazing by marine calanoid copepods (Arthropoda: Crustacea). Science 200: 1403-1405.

Poulet, S.A. and G. Ouellet. 1982. The role of amino acids in the chemosensory swarming and feeding of marine copepods. J. Plank. Res. 4: 341-361.

Poulet, S.A., J.F. Samain, and J. Moal. 1986. Chemoreception, nutrition and food requirements among copepods. Syllogeus 58: 426-442.

Pourriot, R. 1977. Food and feeding habits of Rotifera. Arch. Hydrobiol. Beih. 8: 243-260.

Prepas, E.E. and A.M. Trimbee. 1988. Impact of nitrogen and phosphorous additions on phytoplankton and zooplankton in deep prairie lake. Verh. Internat. Verein. Limnol. 23: 227.

Prescott, G.W. 1979. How to Know the Freshwater Algae. Wm. C. Brown Co., Dubuque. 293pp.

Price, H.J. and G.-A. Paffenhöfer. 1986. Capture of small cells by the copepod *Eucalanus elongatus*. Limnol. Oceanogr. 31: 189-194.

Price, H.J., G.-A. Paffenhöfer, and J.R. Strickler. 1983. Modes of cell capture in calanoid copepods. Limnol. Oceanogr. 28: 116-123.

Quigley, M.A., J.F. Cavaletto, and W.S. Gardner. 1989. Lipid composition related to size and maturity of the amphipod *Pontoporeia hoyi*. J. Great Lakes Res. 15: 601-610.

Ranta, E. and V. Nuutinen. 1985. Foraging by the smooth newt (*Triturus vulgaris*) on zooplankton: functional responses and diet choice. J. Animal Ecology 54: 275-293.

Rao, G.A., D.E. Riley, and E.C. Larkin. 1985. Comparison of the Thin Layer Chromatography/Flame Ionization Detection system with other methods for the quantitative analysis of liver lipid contents in alcohol-fed rats and controls. Lipids 20: 531-535.

Reynolds, C.S. 1984. The Ecology of Freshwater Phytoplankton. Cambridge Univ. Press. 384pp.

Rezeg, T.A. and C.M. James. 1987. Production and nutritional quality of the rotifer *Brachionus plicatilis* fed marine *Chlorella* sp. at different cell densities. Hydrobiologia 147: 257-261.

Reznick, D. 1983. The structure of guppy life histories: the tradeoff between growth and reproduction. Ecology 64: 862-873.

Rice, W.R. and G.W. Salt. 1988. Speciation via disruptive selection on habitat preference: experimental evidence. Am. Nat. 131: 911-917.

Richerson, P., R. Armstrong, and C.R. Goldman. 1970. Contemporaneous disequilibrium, a new hypothesis to explain the "paradox of the plankton". Proc. Nat. Acad. Sci. 67: 1710-1714.

Richman, S. 1966. The effect of phytoplankton concentration of the feeding rate of *Diaptomus oregonensis*. Verh. Internat. Verein. Limnol. 16: 392-398.

Richman, S., S.A. Bohon, and S.E. Robbins. 1980. Grazing interactions among freshwater calanoid copepods. ASLO Spec. Symp. 3: 219-233.

Roman, M.R., K.A. Ashton, and A.L. Gauzens. 1988. Day/night differences in the grazing impact of marine copepods. Hydrobiologia 167/168: 21-30.

Runge, J.A. 1984. Egg production of the marine planktonic copepod, *Calanus pacificus* Brodsky: Laboratory observations. J. Exp. Mar. Biol. Ecol. 74: 53-66.

Sargent, J.R. and S. Falk-Petersen. 1988. The lipid biochemistry of calanoid copepods. Hydrobiologia 167/168: 101-114.

Sargent, J.R. and R.J. Henderson. 1986. Lipids. *In*: E.D.S. Corner and S.C.M. O'Hara (eds.) The Biological Chemistry of Marine Copepods. Clarendon Press, Oxford. pp.59-164.

Sargent, J.R., R.R. Gatten, amd R. McIntosh. 1977. Wax esters in the marine environment -

their occurence, formation, transformation and ultimate fates. Mar. Chem. 5: 573-584.

Scavia, D., G.L. Fahnenstiel, J.A. Davis, and R.G. Kreis, Jr. 1984. Small-scale nutrient patchiness: Some consequences and a new encounter mechanism. Limnol. Oceanogr. 29: 785-793.

Schemmel, R. 1976. Physiological considerations of lipid storage and utilization. Amer. Zool. 16: 661-670.

Schindler, D.W. 1977. Evolution of phosphorous limitation in lakes. Science 195: 260-262.

Schindler, D.W. and E.J. Fee. 1974. Experimental Lakes Area: Whole-lake experiments in eutrophication. Can. J. Fish. Aquat. Sci. 31: 937-953.

Schindler, D.W., H. Kling, R. Schmidt, J. Prokipowich, V. Frost, R. Reid, and M. Capel. 1973. Eutrophication of Lake 227 by addition of phosphate and nitrate: the second, third, and fourth years of enrichment, 1970, 1971, and 1972. Can. J. Fish. Aquat. Sci. 30: 1415-1440.

Schoener, T.W. 1989. Food webs from the small to the large. Ecology 70: 1559-1589.

Scudder, G.G.E. 1974. Species concepts and speciation. Can. J. Zool. 52: 1121-1134.

Smith, A.E. and I. Morris. 1980. Synthesis of lipid during photosynthesis by phytoplankton of the Southern Ocean. Science 207: 197-198.

Smith, S.L. and P.V.Z. Lane. 1985. Laboratory studies of the marine copepod *Centropages typicus*: egg production and development rates. Mar. Biol. 85: 153-162.

Snell, T.W. and C.A. Hawkins. 1983. Behavioral reproductive isolation among populations of the rotifer *Brachionus plicatilis*. Evolution 37: 1294-1305.

Sokal, R.R. and F.J. Rohlf. 1969. Biometry. W.H. Freman & Co., San Francisco. 776pp.

Solomon, M.E. 1949. The natural control of animal populations. J. Anim. Ecology 18: 1-35.

Sommer, U., Z.M. Gliwicz, W. Lampert, and A. Duncan. 1986. The PEG-Model of seasonal succession of planktonic events in fresh waters. Arch. Hydrobiol. 106: 433-471.

Sprules, W.G. and R. Knoechel. 1984. Lake ecosystem dynamics based on functional representations of trophic components. AAAS Sel. Symp. 85: 383-403.

Stearns, S.C. 1989. The evolutionary significance of phenotypic plasticity. BioScience 39: 436-445.

Stein, J.R. 1975. Freshwater algae of British Columbia: the lower Fraser Valley. Syesis 8: 119-184.

Stemberger, R.S. 1986. The effects of food deprivation, prey density, and volume on clearance rates and ingestion rates of *Diacyclops thomasi*. J. Plankton Res. 8: 243-251.

Stephens, D.W. and J.R. Krebs. 1986. Foraging Theory. Princeton Univ. Press, N.J. 247pp.

Stockner, J.G. and K.S. Shortreed. 1985. Whole-lake fertilization experiments in coastal British Columbia lakes: empirical relationships between nutrient inputs and phytoplankton biomass and production. Can. J. Fish. Aquat. Sci. 42: 649-658.

Strickler, J.R. 1982. Calanoid copepods, feeding currents, and the role of gravity. Science 218: 158-160.

Strickler, J.R. 1985. Feeding currents in calanoid copepods: two new hypotheses. *In*: M.S. Lauerack (ed.) Physiological Adaptations of Marine Animals, Symp. Soc. Exp. Biol. 39: 459-485.

Sullivan, B.K. and L.T. McManus. 1986. Factors controlling seasonal succession of the copepods *Acartia hudsonica* and *A. tonsa* in Narragansett Bay, Rhode Island: temperature and resting egg production. Mar. Ecol. Prog. Ser. 28: 121-128.

Suttle, C.A. and P.J. Harrison. 1988a. Ammonium and phosphate uptake rates, N:P supply ratios, and evidence for N and P limitation in some oligotrophic lakes. Limnol. Oceanogr. 33: 186-202.

Suttle, C.A. and P.J. Harrison. 1988b. Rapid ammonium uptake by freshwater phytoplankton. J. Phycol. 24: 13-16.

Suttle, C.A., J.G. Stockner, and P.J. Harrison. 1987. Effects of nutrient pulses on community structure and cell size of a freshwater phytoplankton assemblage in culture. Can. J. Fish. Aquat. Sci. 44: 1768-1774.

Sweeney, B.W., R.L. Vannote, and P.J. Dodds. 1986. Effects of temperature and food quality on growth and development of a mayfly, *Leptophlebia intermedia*. Can. J. Fish. Aquat. Sci. 43: 12-18.

Swift, M.C. and A.Y. Fedorenko. 1975. Some aspects of prey capture by *Chaoborus* larvae. Limnol. Oceanogr. 20: 418-425.

Taber, R.D. 1956. Deer nutrition and population dynamics in the North Coast Range of California. Trans. North Amer. Wildl. Conf. 21: 159-172.

Tauber, C.A. and M.J. Tauber. 1981. Insect seasonal cycles: genetics and evolution. Ann. Rev. Ecol. Syst. 12: 281-308.

Tauber, C.A. and M.J. Tauber. 1989. Sympatric speciation in insects: Perception and perspective. pp. 307-344. *In*: D. Otte and J.A. Endler (eds.) Speciation and Its Consequences. Sinaeur Assoc., Inc. Sunderland, MA.

Templeton, A.R. 1989. The meaning of species and speciation: A genetic perspective. pp. 3-27. *In*: D. Otte and J.A. Endler (eds.) Speciation and Its Consequences. Sinaeur Assoc., Inc. Sunderland, MA.

Tessier, A.J. and C.E. Goulden. 1982. Estimating food limitations in cladoceran populations. Limnol. Oceanogr. 27: 707-717.

Tessier, A.J., L.L. Henry, C.E. Goulden, and M.W. Durand. 1983. Starvation in *Daphnia*: Energy reserves and reproductive allocation. Limnol. Oceanogr. 28: 667-676.

Thompson, D.J. 1986. Heritability for body size in the isopod *Asellus aquaticus* (L.). Crustaceana 51: 241-244.

Threlkeld, S.T. 1986. Differential temperature sensitivity of two cladoceran species to resource variation during a blue-green algal bloom. Can. J. Zool. 64: 1739-1744.

Threlkeld, S.T. and J.M. Dirnberger. 1986. Benthic distributions of planktonic copepods,

especially *Mesocyclops edax*. Syllogeus 58: 481-486.

Tilman, D. 1981. Tests of resource competition theory using four species of Lake Michigan algae. Ecology 62: 802-825.

Tilman, D. 1982. Resource Competition and Community Structure. Monographs in Population Biology, Princeton. 296pp.

Tilman, D., R. Kiesling, R. Sterner, S.S. Kilham, and F.A. Johnson. 1986. Green, bluegreen and diatom algae - taxonomic differences in competitive ability for phosphorous, silicon and nitrogen. Arch. Hydrobiol. 106: 473-485.

Tinbergen, L. 1960. The natural control of insects in pinewoods: I: Factors influencing the intensity of predation by songbirds. Arch. Néerl. Zool. 13: 266-336.

Turpin, D.H. 1986. Growth rate dependent optimum ratios in *Selenastrum minutum* (Chlorophyta): implications for competition, coexistence and stability in phytoplankton communities. J. Phycol. 22: 94-102.

Turpin, D.H. and P.J. Harrison. 1980. Cell size manipulation in natural marine, planktonic, diatom communities. Can. J. Fish. Aquat. Sci. 37: 1193-1195.

Van Alstyne, K.L. 1986. Effects of phytoplankton taste and smell on feeding behavior of the copepod *Centropages hamatus*. Mar. Ecol. Prog. Ser. 34: 187-190.

Vanderploeg, H.A. 1981. Seasonal particle-size selection by *Diaptomus sicilis* in offshore Lake Michigan. Can. J. Fish. Aquat. Sci. 38: 504-517.

Vanderploeg, H.A. 1990. Feeding mechanisms and their relation to particle selection and feeding in suspension-feeding zooplankton. *In*: R.S. Wotton (ed.) The Biology of Particles in Aquatic Systems. CRC Press, Boca Raton, FL. 320pp.

Vanderploeg, H.A. and G.-A. Paffenhöfer. 1985. Modes of algal capture by the freshwater copepod *Diaptomus sicilis* and their relation to food-size selection. Limnol. Oceanogr. 30: 871-885.

Vanderploeg, H.A. and D. Scavia. 1979. Calculation and use of selectivity coefficients of feeding: zooplankton grazing. Ecol. Model. 7: 135-149.

Vanderploeg, H.A., G.-A. Paffenhöfer, and J.R. Liebig. 1989. Concentration-variable interactions between calanoid copepods and particles of different food quality: observations and hypotheses. *In*: (R.N. Hughes, ed.) Behavioral Mechanisms of Food Selection. NATO ASI Series, Series A: Life Sciences, Springer Verlag, Heidelberg.

Vanderploeg, H.A. , D. Scavia, and J.R. Liebig. 1984. Feeding rate of *Diaptomus sicilis* and its relation to selectivity and effective food concentration in algal mixtures and in Lake Michigan. J. Plank. Res. 6: 919-941.

Vanni, M.J. 1986. Competition in zooplankton communities: Suppression of small species by *Daphnia pulex*. Limnol. Oceanogr. 31: 1039-1056.

Vanni, M.J. 1987. Effects of food availability and fish predation on a zooplankton community. Ecol. Monogr. 57: 61-88.

Vanni, M.J. and D.L. Findlay. 1990. Trophic cascades and phytoplankton community structure. Ecology 71: 921-937.

Venrick, E.L. 1990. Phytoplankton in an oligotrophic ocean: species structure and interannual variability. Ecology 71: 1547-1563.

Verity, P.G. and T.J. Smayda. 1989. Nutritional value of *Phaeocystis pouchetii* (Prymnesiophyceae) and other phytoplankton for *Acartia* spp. (Copepoda): ingestion, egg production, and growth of nauplii. Mar. Biol. 100: 161-171.

Vollenweider, R.A. 1976. Advances in defining critical loading levels for phosphorous in lake eutrophication. Mem. Ist. Ital. Idrobiol. 33: 53-83.

Walters, C.J., E. Krause, W.E. Neill, and T.G. Northcote. 1987. Equilibrium models for seasonal dynamics of plankton biomass in four oligotrophic lakes. Can. J. Fish. Aquat. Sci. 44: 1002-1017.

Walters, C.J., D.C.E. Robinson, and T.G. Northcote. 1990. Comparative population dynamics of *Daphnia rosea* and *Holopedium gibberum* in four oligotrophic lakes. Can. J. Fish. Aquat. Sci. 47: 401-409.

Warren, G.J., M.S. Evans, D.J. Jude, and J.C. Ayers. 1986. Seasonal variations in copepod size: effects of temperature, food abundance, and vertebrate predation. J. Plank. Res. 8: 841-853.

Werring, J. 1986. Fertilization of an oligotrophic coastal montane lake using frequent doses of high N:P ratio fertilizers: Effects on phytoplankton and zooplankton community structure. M.Sci. Thesis, U.B.C. 295pp.

Wetzel, R.G. 1983. Limnology. Saunders College Publishing, Philadelphia. 860pp.

Whyte, J.N.C., J.R. Englar, B.L. Carswell, and K.E. Medic. 1986. Influence of starvation and subsequent feeding on body composition and energy reserves in the prawn *Pandalus platyceros*. Can. J. Fish. Aquat. Sci. 43: 1142-1148.

Williamson, C.E. 1987. Predator-prey interactions between omnivorous diaptomid copepods and rotifers: The role of prey morphology and behavior. Limnol. Oceanogr. 32: 167-177.

Williamson, C.E. and N.M. Butler. 1986. Predation on rotifers by the suspension-feeding calanoid copepod *Diaptomus pallidus*. Limnol. Oceanogr. 31: 393-402.

Williamson, C.E. and N.M. Butler. 1987. Temperature, food and mate limitation of copepod reproductive rates: separating the effects of multiple hypotheses. J. Plank. Res. 9: 821-836.

Williamson, C.E. and H.A. Vanderploeg. 1988. Predatory suspension-feeding in *Diaptomus*: prey defenses and the avoidance of cannibalism. Bull. Mar. Sci. 43: 561-572.

Williamson, C.E., N.M. Butler, and L. Forcina. 1985. Food limitations in naupliar and adult *Diaptomus pallidus*. Limnol. Oceanogr. 30: 1283-1290.

Wilson, M.S. 1953. New and inadequately known North American species of the copepod genus *Diaptomus*. Smithsonian Misc. Coll. 122: 1-30.

Woodward, I.O. and R.W.G. White. 1981. Effects of temperature and food on the fecundity and egg development rates of *Boeckella symmetrica* Sars (Copepod: Calanoida). Australian J. Mar. Freshw. Res. 32: 997-1002.

Wurdak, E.S. and J.J. Gilbert. 1980. Ultrastructure and histochemistry of the rudimentary gut of male *Asplanchna sieboldi* (Rotifera). Hydrobiologia 73: 123-126.

Wyngaard, G.A. 1986. Genetic differentiation of life history traits in populations of *Mesocyclops edax* (Crustacea: Copepoda). Biol. Bull. 170: 279-295.

Zaret, T.M. 1980. Predation and Freshwater Communities. Yale Univ. Press, New Haven.