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INVERTEBRATE TROPHIC RELATIONSHIPS
IN TEMPORARY WOODLAND PONDS IN MICHIGAN

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INVERTEBRATE TROPHIC RELATIONSHIPS IN TEMPORARY WOODLAND PONDS IN MICHIGAN

By

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ABSTRACT

INVERTEBRATE TROPHIC RELATIONSHIPS INTEMPORARY WOODLAND PONDS IN MICHIGAN

By

Michael John Higgins

Temporary woodland ponds are small, fishless habitats that flood in the early spring from snowmelt and rainfall. They may remain flooded for 8-50 weeks out of the year, depending upon the size of the pond. While much of the energy flow within these systems is detritus based, primary production is also very important in medium to large-sized ponds. The invertebrate communities that inhabit these ponds are all characterized by rapid development and possess either a drought-resistant stage or are capable of dispersal to permanent water when the habitat dries. Mosquito larvae are the most abundant insects within these habitats, and the feeding ecology of larval *Aedes stimulans* mosquitoes was examined through field microcosm experiments. Larvae of mosquitoes with little material on which to graze survived and grew as well as those furnished with a biofilm on which they could feed. Analysis of gut contents indicated that the non-grazing mosquitoes filtered substantially more high quality algae from the water column than the grazing mosquito larvae, suggesting that the algae was an important component of their diet. Other studies indicate that the invertebrate communities in these temporary ponds are strongly influenced by sized-based predation, and that phenology may be important for many prey species. Potential prey organisms such as mosquitoes, fairy shrimp, and cladocerans may reduce their exposure to predation by beginning their development early in the spring prior to the appearance of most predators. In addition, by beginning development early they can reach a body size that is beyond the handling capabilities of most later-appearing predators. Analysis of the cladoceran community indicates seasonal shifts in species composition, body size, and morphology that correspond to seasonal changes in the composition of the predator community.

This dissertation is dedicated to the memory of Robert and Mary Ann,
beloved father and sister.

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A Ph.D. dissertation, being the culmination of one's academic training, is, in many respects, the result of one's knowledge and experiences gained in life up to that point. So many people and events throughout our lives have forged our way of thinking in such subtle ways that we could never remember them all, much less acknowledge them. And yet, looking back, we can see moments and places where paths diverged and one's life turned to follow one over the other. More often than not, our choices were guided by individuals who showed us ways of viewing life that we had not before considered; individuals who—in retrospect—helped shape our lives. Science, like life itself, builds on what has come before. The dissertation that follows is the direct result of my five years of study at Michigan State University, but it is also the fruit of influences past and present.

I would first like to thank my parents, Norma and Robert for their support over the years. My undergraduate mentor in anthropology, Dr. Richard Flanders taught me how to think critically. At Western Michigan University, special thanks go to Drs. William Cremin and Betsy Garland of the Department of Anthropology, and Drs. Richard Brewer and David Cowan of the Department of Biological Sciences. Dr. Brewer provided me with a strong foundation in ecology, and Dr. Cowan was responsible for instilling in me a passion for entomology and aquatic invertebrates. Through his courses in evolutionary biology, Dr. Cowan also started me to think as a biologist.

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INTRODUCTION

Temporary woodland ponds are small seasonal wetlands that may hold water from 2-9 months out of the year. They may vary in size from less than 10 m^2 to over 0.5 ha, but most are probably less than 0.1 ha. These unique habitats have received relatively little attention in the scientific literature and--due their small size--practically no protection from human impact. In southern Michigan forests and woodlots, the remaining ponds are no doubt a tiny fraction of number that once dotted the presettlement landscape.

Despite their diminutive size, temporary woodland ponds are exclusive breeding sites for a number of vertebrate and invertebrate organisms. Although these ponds generally do not support waterfowl, a number of amphibians use these seasonal habitats as breeding sites. In particular, Wood Frogs, *Rana sylvatica*, Western Chorus Frogs, *Pseudacris triseriata triseriata*, and several species of mole salamanders, *Ambystoma* spp., use woodland pools for breeding and larval development in the spring (Harding 1997). In addition, Gray Treefrogs, *Hyla versicolor*, and Spring Peepers, *Pseudacris crucifer crucifer*, may also use these small wetlands for breeding purposes (Harding 1997).

Several invertebrate species are unique to temporary ponds, and woodland ponds in particular. Fairy shrimp (Family Chirocephalidae) occur only in temporary ponds, and several species of *Eubranchipus* are common in woodland pools in the eastern United States. The cladoceran, *Daphnia ephemeralis*, described as a species only in 1985 (Schwartz and Hebert 1985), is apparently only found in temporary woodland ponds in eastern North America. Another cladoceran, *Simocephalus exspinosus*, may also favor temporary habitats. Several species of insects are adapted to temporary ponds, and are found exclusively in these seasonal habitats. The damselfly, *Lestes dryas*, oviposits in plant tissue along the margins of dry woodland pools, and *Sympetrum* dragonflies (especially *obtrusum* and *rubicundulum*) also oviposit in the dry basins. These odonates undergo obligate diapause as eggs and do not hatch until the following season (Wiggins et al. 1980). At least one species of beetle, the dytiscid *Agabus erichsoni*, is particularly well adapted to temporary ponds, diapausing as eggs in pond basins during the dry phase. Similarly, larvae of the phantom midge, *Mochlonyx*, are found exclusively in temporary ponds.

In terms of abundance and impact on humans, mosquito larvae are perhaps the most important organism to inhabit temporary ponds. Several species in the genus *Aedes* (in particular, *excucians*, *provocans*, and *stimulans*) are very common in temporary woodland ponds and, in many ways, epitomize an organism with the unique adaptations required for survival and growth in these ephemeral habitats. *Aedes* mosquitoes have drought and freeze-tolerant eggs that may remain viable in a suitable habitat for several years(Horsfall 1956). Larvae can survive and feed at temperatures of 0° C during the late winter and early spring (Westwood et al. 1983; Walker 1995), and some species can develop from egg to adult in as little as 5 days in summer rain pools (Horsfall 1955). In addition to *Aedes*, several species of *Psorophora* mosquitoes are common in summer rain pools in the eastern North America (Horsfall 1955; Wood et al. 1979). Two species in particular, *P. ciliata* and *P. ferox*, are common in woodland rain pools in Michigan during the summer.

In addition to being a major nuisance to humans engaged in outdoor activities, many of the mosquitoes that breed in temporary woodland ponds are vectors of diseases that afflict humans and animals. Several *Aedes* mosquitoes are competent vectors of the California encephalitis group of viruses (LaCrosse and Jamestown Canyon), and a few--most notably, *A. vexans*-- are capable of transmitting Eastern and/or Western Equine encephalitis (Harwood and James 1979). *Psorophora ciliata* is a competent vector for St. Louis encephalitis. Several species of *Aedes* mosquitoes are also potential vectors of dog heartworm (Ludlam et al. 1970).

Despite the unique organisms and the potential disease vectors that inhabit small temporary ponds, these habitats have--until only recently--received relatively little scientific scrutiny. Mozley (1928, 1932) published one of the first descriptive studies of a temporary pond and its inhabitants, and Kenk (1949) compared temporary and permanent pond communities in southern Michigan. In the 1950s and 1960s, there were studies that examined the ecology of some *Aedes* mosquito larvae (e.g., Horsfall 1956; Haufe and Burgess 1956; Haufe 1957; Horsfall and Fowler 1961; James 1966), and potential predators of mosquito larvae in snowmelt pools (Baldwin et al. 1955; James 1961, 1969). There have been studies of the population ecology of temporary pond Aedes mosquitoes (Iversen 1971; Enfield and Pritchard 1977) and a few studies examining the feeding ecology of these mosquitoes (Hinman 1930; Howland 1930; Ameen and Iversen 1978). Barlocher et al. (1978) examined the importance of the dry phase of temporary ponds in conditioning the leaf detritus. They determined that leaves that were colonized by terrestrial fungi provided a more nutritive food source for detritvorous insects than leaves that remained submerged for the same length of time.

More recently, there have been several syntheses regarding the ecology of temporary ponds (Williams 1987, 1996) and wetlands in general, including temporary ponds (Batzer and Wissinger 1996; Batzer et al. 1999).

In a landmark paper, Wiggins et al (1980) examined the adaptive strategies of animals inhabiting temporary ponds. Four strategies were recognized based on physical adaptations to survive the dry phase and oviposition behavior. Group 1 inhabitants are permanent residents possessing either a drought-resistant stage or the ability to burrow into moist sediments. Dispersal is generally passive only. Group 2 inhabitants overwinter in some drought-resistant stage, emerge as adults in the spring, and oviposit in the pond before it dries. Group 3 is similar to Group 2 except that oviposition is independent of the presence of water, so oviposition generally occurs in the dry basin. Group 4 individuals possess no particular adaptation to the dry phase and must recolonize the pond each spring. Larvae of Group 4 inhabitants must complete development and emigrate to a permanent water source before the pond dries. The description of these 4 strategies by Wiggins et al. (1980) still provides a very useful paradigm for viewing the adaptations of animals to life in ephemeral habitats.

Beyond the abiotic constraints placed on inhabitants of ephemeral ponds, biotic interactions are also important in structuring communities. Recently, the role of predation in shaping temporary pond communities has been examined (Wellborn et al. 1996; Schneider and Frost 1996; Schneider 1997, 1999), particularly with regard to pond duration. Long-duration ponds support a much more diverse and, on average, longer-lived predator community than do short duration ponds. Such predator communities may exclude certain taxa from long-duration ponds, or limit population sizes of other taxa (Schneider and Frost 1996). The mechanisms for such exclusion or limitation may include behavioral tradeoffs (Wellborn et al. 1996). Animals in temporary habitats need to grow quickly and thus forage actively. This foraging activity, however, results in increased exposure to predation. In short-duration ponds with limited predator communities, this may not present a problem. In long-duration and permanent ponds, however, such active foraging may lead to extermination by predators. Thus, animals that are well adapted to temporary ponds are often excluded from permanent and semi-permanent ponds (Wellborn et al. 1996). Such mechanisms are viewed as accessory to life history constraints such as a prerequisite cold and dry period to initiate egg hatching (e.g., most *Aedes* mosquitoes). That many permanent and semi-permanent pond communities contain many temporary pond taxa and a reduced predator community after a drought (Jeffries 1994; Schneider and Frost 1996) supports the argument that such taxa are excluded by predation.

The research presented in the chapters that follow represents the results of a multi-year study of the invertebrate communities in several temporary woodland ponds in the Lansing, Michigan area. These ponds ranged in size from 5 m^2-3700 m^2, and from 10-50 weeks in duration. Chapter 1 presents an overview of temporary woodland ponds in Michigan and a summary of some of the research. Chapter 2 examines the feeding ecology of *Aedes stimulans* larvae in these ponds. Chapter 3 describes the zooplankton community and cladoceran succession through the wet phase of one woodland pond. Chapter 4 examines predator-prey relationships and the role of both phenology and body size.

REFERENCES

Ameen, M. and T. M. Iversen. 1978. Food of *Aedes* larvae (Diptera: Culicidae) in a temporary forest pool. Archiv für Hydrobiologie 83: 552-564.

Baldwin, W. F., H. G. James, and H. E. Welch. 1955. A study of mosquito larvae and pupae with a radio-active tracer. Canadian Entomologist 87: 350-356.

Barlocher, F., R. J. Mackay, and G. B. Wiggins. 1978. Detritus processing in a temporary vernal pool in southern Ontario. Archiv für Hydrobiologie 81: 269-295.

Batzer, D. P., Rader, R. B., and Wissinger, S. A. [eds.] 1999. Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley and Sons. New York.

Batzer, D. P. and S. A. Wissinger. 1996. Ecology of insect communities in nontidal wetlands. Annual Review of Entomology 41: 75-100.

Enfield, M. A. and G. Pritchard. 1977. Estimates of population size and survival of immature stages of four species of *Aedes* (Diptera: Culicidae) in a temporary pond. Canadian Entomologist 109: 1425-1434.

Harding, J. H. 1997. Amphibians and Reptiles of the Great Lakes Region. University of Michigan Press. Ann Arbor.

Harwood, R. F. and M. T. James. 1979. Entomology in Human and Animal Health, 7th. ed. Macmillan. New York.

Haufe, W. O. 1957. Physical environment and behavior of immature stages of *Aedes communis* (Deg.) in subarctic Canada. Canadian Entomologist 89: 120-139.

Haufe, W. O. and L. Burgess. 1956. Development of Aedes (Diptera: Culicidae) at Fort Churchill, Manitoba, and prediction of dates of emergence. Ecology 37: 500-519.

Hinman, E. H. 1930. A study of the food of mosquito larvae (Culicidae). American Journal of Hygiene 12: 238-270.

Horsfall, W. R. 1955. Mosquitoes: Their Bionomics and Relation to Disease. Ronald Press. New York.

Horsfall, W. R. 1956. Eggs of floodwater mosquitoes (Diptera: Culicidae). III. Condition and hatching of *Aedes vexans*. Annals of the Entomological Society of America 49: 66-71.

Horsfall, W. R. and H. W. Jr. Fowler. 1961. Eggs of floodwater mosquitoes VIII. Effect of serial temperatures on conditioning of eggs of *Aedes stimulans* Walker (Diptera: Culicidae). Annals of the Entomological Society of America 54: 664-666.

Howland, L. J. 1930. Bionomical investigation of English mosquito larvae with special reference to their algal food. Journal of Ecology 18: 81-125.

Iversen, T. M. 1971. The ecology of a mosquito population (*Aedes communis*) in a temporary pool in a Danish beech wood. Archiv für Hydrobiologie 69: 309-332.

James, H. G. 1961. Some predators of *Aedes stimulans* (Walk.) and *Aedes trichurus* (Dyar) (Diptera: Culicidae) in woodland pools. Canadian Journal of Zoology - Journal Canadien de Zoologie 39: 533-540.

James, H. G. 1966. Location of univoltine Aedes eggs in woodland pool areas and experimental exposure to predators. Mosquito News 26: 59-63.

James, H. G. 1969. Immature stages of five diving beetles (Coleoptera: Dytiscidae), notes on their habits and life history, and a key to aquatic beetles of vernal woodland pools in southern Ontario. Proceedings of the Entomological Society of Ontario 100: 52-97.

Jeffries, M. 1994. Invertebrate communities and turnover in wetland ponds affected by drought. Freshwater Biology 32: 603-612.

Kenk, R. The animal life of temporary and permanent ponds in southern Michigan. (71). 1949. University of Michigan, Museum of Zoology. Miscellaneous Publication.
Ref Type: Serial (Book,Monograph)

Ludlam, K. W., L. A. Jr. Jachowski, and G. F. Otto. 1970. Potential vectors of *Dirofilaria immitis*. Journal of the American Veterinary Medical Association 157: 1354-1359.

Mozley, A. 1928. Note on some fresh water mollusca inhabiting temporary ponds in western Canada. Nautilus 42: 19-20.

Mozley, A. 1932. A biological study of a temporary pond in western Canada. American Midland Naturalist 66: 235-249.

Schneider, D. W. 1997. Predation and food web structure along a habitat duration gradient. Oecologia 110: 567-575.

Schneider, D. W. 1999. Snowmelt ponds in Wisconsin: Influence of hydroperiod on invertebrate community structure, p. 299-318. *In* [eds.], D.P.Batzer, R.B.Rader, and S.A.Wissinger, Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley and Sons, New York.

Schneider, D. W. and T. M. Frost. 1996. Habitat duration and community structure in temporary ponds. Journal of the North American Benthological Society 15: 64-86.

Schwartz, S. S. and P. D. N. Hebert. 1985. *Daphniopsis ephemeralis* sp.n. (Cladocera, Daphniidae): a new genus for North America. Canadian Journal of Zoology - Journal Canadien de Zoologie 63: 2689-2693.

Walker, E. D. 1995. Effect of low temperature on feeding rate of *Aedes stimulans* larvae and efficacy of *Bacillus thuringiensis* var. *israelensis* (H-14). Journal of the American Mosquito Control Association 11: 107-110.

Wellborn, G. A., D. K. Skelly, and E. E. Werner. 1996. Mechanisms creating community structure across a freshwater habitat gradient. Annual Review of Ecology and Systematics 27: 337-363.

Westwood, A. R., G. A. Surgeoner, and B. V. Helson. 1983. Survival of spring *Aedes* spp mosquito (Diptera: Culicidae) larvae in ice-covered pools. Canadian Entomologist 115: 195-197.

Wiggins, G. B., R. J. Mackay, and I. Smith. 1980. Evolutionary and ecological strategies of animals in annual temporary pools. Archiv für Hydrobiologie, Supplement 58: 97-206.

Williams, D. D. 1987. The Ecology of Temporary Waters. Croom Helm, Timber Press. Portland, OR.

Williams, D. D. 1996. Environmental constraints in temporary fresh water and their consequences for the insect fauna. Journal of the North American Benthological Society 15: 634-650.

Wood, D. M., Dang, P. T., and Ellis, R. A. 1979. The mosquitoes of Canada (Diptera: Culicidae). The Insects and Arachnids of Canada, Part 6. Publication 1686. Ottawa, Ontario, Canada, Biosystematics Research Institute, Research Branch, Agriculture Canada.

CHAPTER 1

TEMPORARY WOODLAND PONDS IN MICHIGAN: INVERTEBRATE SEASONAL PATTERNS AND TROPHIC RELATIONSHIPS

ABSTRACT

Temporary woodland ponds are relatively small, shallow wetlands that retain water for a few weeks to several months out of the year. Most of the energy flow within these habitats stems from microbial degradation of leaf litter deposited by surrounding trees and shrubs. The composition of the invertebrate community found within any particular pond is related to its size and duration of flooding. The invertebrates that inhabit these ponds show varying degrees of adaptation to ephemeral habitats, but nearly all are characterized by rapid larval growth. Medium to large size ponds exhibit a predictable seasonal succession of species, a pattern that has evolved in response to both physical constraints and biotic interactions. The early-season inhabitants are particularly well-adapted to both the ephemeral habitat as well as the cold temperatures characteristic of early spring in Michigan. These animals feed primarily on the abundant microbial community present on the the leaf litter and within the water column, and avoid heavy predation pressure by beginning development before the appearance of most of the predators. Most of the ponds' inhabitants that are not specifically adapted to ephemeral habitats are predators. These are generally insects that overwinter in permanent water and recolonize temporary ponds each spring. By consuming a high-quality food source such as animal protein, these migrants are able to develop rapidly and thus ensure completion of the larval phase before the ponds dry.

INTRODUCTION

Prior to Euro-American settlement, the wooded landscape of southern Michigan was dotted by innumerable ephemeral ponds formed millennia ago in vast glacial outwash plains. Although only a small percentage of these temporary woodland ponds remain today, they represent a fairly common, yet remarkably understudied aquatic habitat. Woodland ponds can range in size from a few square meters to over a hectare, although most are probably less than 0.4 ha. Maximum water depth is generally less than 1.5 m, and the average depth is usually under 1 meter. These small woodland ponds are generally unsuitable for waterfowl production compared to larger, more open habitats, a factor which may explain the relative inattention these wetlands have received in the scientific literature. Ponds may begin to flood in late autumn or winter, reaching maximum size in the early spring as a result of snowmelt and spring rains. A distinction has been made between vernal pools, which flood only in the spring, and autumnal ponds, which flood in the autumn and remain wet until the following summer (Wiggins et al. 1980). It should be pointed out that the flooding which occurs in autumn often only covers the deepest parts of these ponds, and much of the pond area remains dry until the following spring. While the flooded area of an autumnal pond may provide important overwintering habitat for some aquatic invertebrates that lack specific adaptations for drying and freezing (Batzer and Sion 1999), much of the well-adapted temporary pond fauna remains unaffected by this flood event. Thus, while the presence or absence of water during the autumn and winter will influence faunal composition somewhat (Kenk 1949; Wiggins et al. 1980; Batzer and Sion 1999), we believe that the size and duration of a particular pond during the vernal phase has an even more pronounced influence on community composition.

The duration of flooding is directly related to area and depth. All of these water bodies are closed depressions and dry from evaporation and groundwater outflow. Most are dry by the middle of summer and some may undergo a second, somewhat accelerated cycle of flooding and drying in the mid to late summer as a result of heavy precipitation from thunderstorms. Flooding during this aestival (summer) phase is generally smaller in areal extent compared with the vernal phase. Small vernal pools flood only in the spring.

Temporary woodland ponds occur in forested landscapes and are thus bordered on all sides by trees (e.g., red maple, silver maple, elm, cottonwood) and shrubs (e.g., dogwoods, alder, spicebush). Trees frequently occur within the flooded portions of the ponds as well as the borders. Because of the intense shading from trees and shrubs along the margins of these ponds, there is often very little emergent vegetation present, or the emergent plants may occur only in relatively small areas that receive sufficient sunlight. In addition, submerged aquatic vegetation (including submerged macrophytes and mats of filamentous algae) generally does not occur in these ponds. After the trees leaf out in the spring, the ponds themselves may receive little direct sunlight, a factor which may limit algal growth compared to more open types of wetlands (Kenk 1949). The surrounding woodland vegetation is also important for the input of substantial amounts of leaf litter into the dry basins in the fall. Barlocher et al. (1978) recorded an average of 132.8 g/m^2 of leaf litter (ash-free dry weight) falling into Ontario pond basins in the autumn.

In southern Michigan, water temperatures range from 3° C in the early spring when ice is still present, to 27° C in the summer. Because of the shallow nature of these bodies of water, daily water temperature fluctuations of 5° C are not uncommon in the spring, particularly in the small ponds (Figure 1). In addition, a thin layer of ice frequently covers the surface at night during the early spring. Early in the season, pH generally ranges between 7-7.5, and gradually becomes more alkaline (7.5-8) as the ponds shrink in size during the late spring and summer. Due to the large surface-to-volume ratio, ice-free ponds do not often become anoxic, but anaerobic conditions exist in the underlying sediments. Dissolved oxygen and pH also undergo diel fluctuations as a result of increase algal respiration at night (Williams 1987).

SEASONAL PATTERNS—VERNAL PHASE

In a landmark paper on temporary pond ecology, Wiggins et al. (1980) divided temporary pond breeding inhabitants into 4 groups (Table 1), based on their adaptations (or lack thereof) to the dry phase of these habitats and also on their oviposition/colonization habits. Animals that are particularly well-adapted to life in these temporary environments (Groups 1-3) are basically year-round residents, spending the dry phase in some drought-resistant stage (often the egg stage). Organisms which lack drought-resistance (Group 4) must move to permanent water before the ponds dry and then recolonize the temporary ponds the following spring. This latter category is characterized by species with excellent colonizing abilities and rapid larval development.

The composition of the invertebrate community present in any given pond is related to its size and duration of flooding (Schneider and Frost 1996; Schneider 1999). Small ponds of only a few weeks duration contain relatively few species, and are dominated

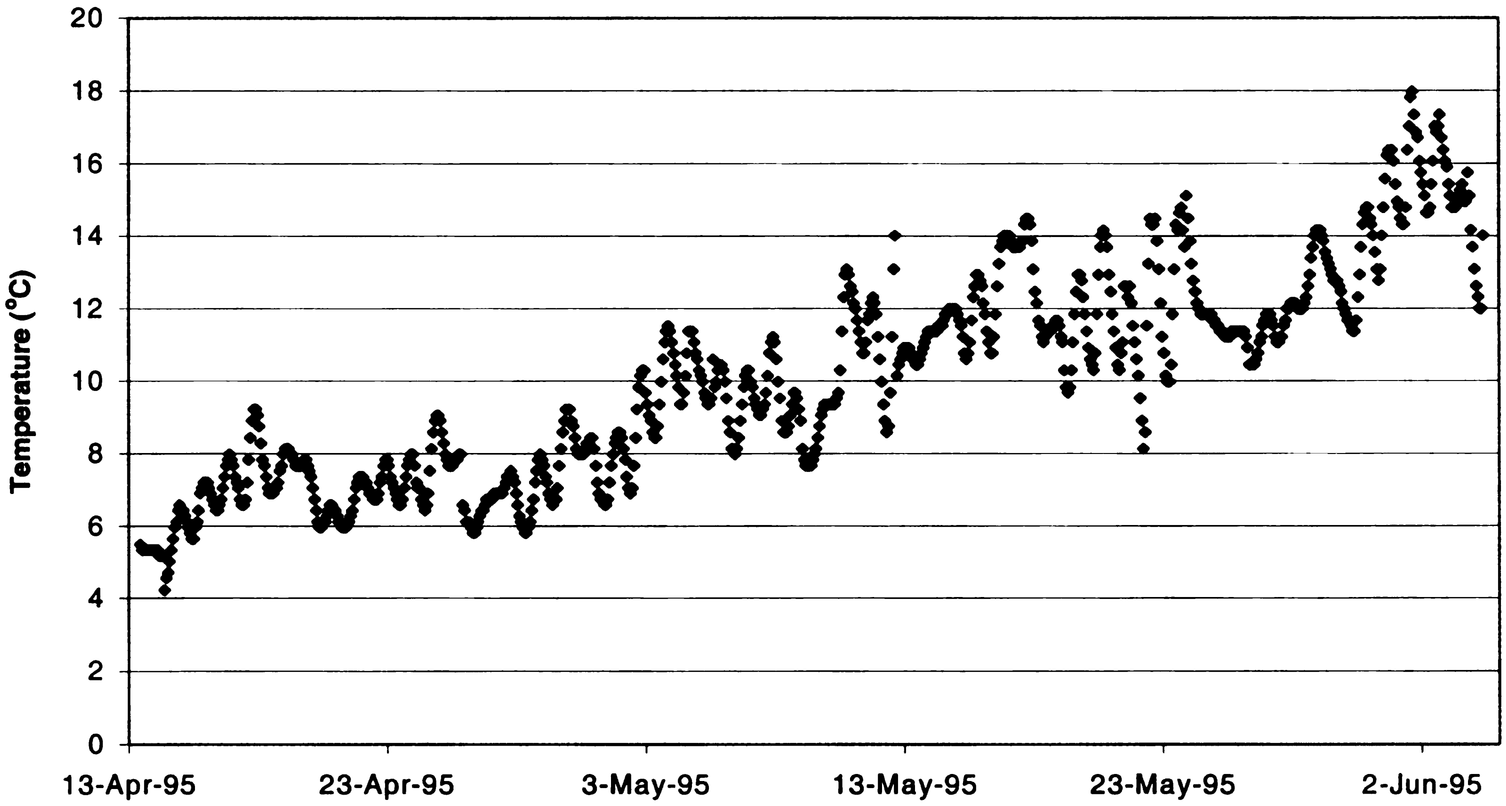

Figure 1. Temperature data for a small vernal pool in southern Michigan. Each symbol represents a reading taken at approximately 1.5 hour intervals with a data logger.

Table 1. Summary of Life History Groups for Invertebrates Inhabiting Temporary Woodland Ponds.

*GROUP**	*GENERAL CHARACTERISTICS*	*EXAMPLES*
1. Overwintering Residents	Permanent residents with drought-resistant stage Passive dispersal only Generally non-insect invertebrates	cladocerans, copepods, ostracods, fairy shrimp, planarians, gastropods, fingernail clams
2. Overwintering Spring Recruits	Nearly permanent residents with drought-resistant stage (usually the egg stage) Adults oviposit in water before pond dries	some dytiscid beetles (e.g., *Agabus*) *Polycentropis crassicornis* caddisfly soldier flies (Statiomyidae)
3. Overwintering Summer Recruits	Nearly permanent residents with drought-resistant stage (usually egg stage) Adults oviposit in dry basin	*Aedes* and *Psorophora* mosquitoes, caddisflies (*Limnephilus*), damselflies (*Lestes*), dragonflies (*Sympetrum*)
4. Non-wintering Spring Migrants	No drought-resistant stage, must re-colonize pond each spring Excellent dispersal capabilities Overwinter in permanent water	all hemipterans (e.g., Corixidae, Gerridae, Notonectidae, Veliidae), some dytiscid beetles (e.g., *Acilius*, *Colymbetes*, *Dyticus*), green darner dragonfly (*Anax junius*)

*from Wiggins et al. 1980

by ostracods and mosquito larvae of the genus *Aedes*. In addition, there may be gastropods, triclad turbellarians (planarians), cladocerans, and occasional predatory beetle larvae (family Dytiscidae) present. Amphipods (*Crangonyx*) were surprisingly abundant in several small vernal pools we have sampled. Amphipods are not thought to be particularly well adapted to the dry and frozen conditions that characterize vernal ponds during the autumn and winter (Wiggins et al. 1980; Batzer and Sion 1999), and their presence in these short-duration (8-10) vernal pools remains an enigma. Isopods, which are also poorly adapted to temporary ponds, were occasionally recovered from a few vernal pools. Their presence may be explained, however, by the co-occurrence of crayfish in these particular ponds. Crayfish burrows may provide a refuge in temporary habitats for poorly adapted organisms such as isopods (Wiggins et al. 1980).

The relatively few number of species present in these small pools suggests relatively simple trophic relationships (Schneider and Frost 1996). Medium- to large-sized ponds, persisting for 4-6 months in the spring and summer, generally contain a large diversity of invertebrate organisms and are characterized by a fairly predictible succession of species. While there may be some invertebrates active during the winter in autumnal ponds--generally small crustaceans such as copepods, ostracods and cladocerans--the primary season of invertebrate activity begins when ice melts along the margins of the ponds, usually in late February or early March in southern Michigan. If the ponds are sufficiently flooded, *Aedes* mosquito eggs that were deposited in the dry basins by females the previous season begin to hatch. Analyses of soil samples from temporary ponds indicate that the vast majority of mosquito eggs are laid near the margins of maximum flooding extent (James 1966) (Iversen 1971). Thus, if insufficient precipitation occurs, resulting in lower than normal water levels, little hatching will take place.

In addition to *Aedes* mosquito larvae, other early-season inhabitants of medium to large-sized temporary woodland ponds include fairy shrimp (order Anostraca), small crustaceans (copepods, cladocerans, ostracods), midge larvae (Chironomidae), phantom midge larvae (genus *Mochlonyx*), caddisfly larvae (primarily *Limnephilus*), and gastropods (e.g., *Physella*). Except for *Mochlonyx*, which preys upon small crustaceans and perhaps some first-instar mosquito larvae, there are few predators during this early part of the season. Conditions at this time are harsh, with cold water temperatures (<10° C) and frequent ice formation on the surface, factors which do not appear to harm the organisms listed above. Mosquito larvae have been shown to survive 10 days of ice cover on a woodland pond in Ontario ((Westwood et al. 1983), and Walker (1995) demonstrated that larvae continue to feed, albeit more slowly, at temperatures down to 0° C. Most of the predators in temporary woodland ponds do not appear until water temperatures exceed 10° C on a regular basis. One of the few predators that appears to have solved physiological problems associated with cold and ice is the dytiscid beetle larva, *Agabus erichsoni*, the eggs of which hatch almost simultaneously with those of mosquitoes. Cold early spring temperatures and short-term ice cover do not seem to have a negative impact on this species, whose primary prey appears to be mosquito larvae (James 1961).

As temperatures warm in mid to late April, more species--chiefly predators--make their appearance within the ponds. Dragonflies and damselflies (*Sympetrum* and *Lestes*) present as eggs laid within the basin the previous summer, hatch out as very small individuals and consume small crustaceans during their early instars. Eggs of a predatory caddisfly, *Polycentropis crassicornis* (Polycentropidae), hatch in late April, and the larvae initially feed on small crustaceans that become trapped in the caddisflies' silken retreats constructed within the leaf litter (Higgins, unpubl. data). Adult water striders (Gerridae) and backswimmers (Notonectidae) that overwintered in permanent water recolonize these temporary environments, and begin breeding at this time, as do several species of beetles (e.g., Dytiscidae: *Acilius*, *Colymbetes*, *Dytiscus*, *Rhantus*; Hydrophilidae: *Hydrochara*, *Hydrochus*, *Tropisternus*; Gyrinidae: *Gyrinus*). Eggs of chorus frogs (*Pseudacris triseriata*) and salamanders (*Ambystoma*) that were laid in the ponds in late March or early April hatch by late April. *Ambystoma* larvae are the only important vertebrate predators in temporary woodland ponds.

Adult mosquitoes begin to emerge from the ponds in early to mid-May of most years. By this same time, fairy shrimp have completed development, deposited eggs, and died. Limnephilid caddisflies, which have been feeding continuously on leaf detritus for two months, reach their final instar by the middle of the month and begin pupating in late May. With the emergence of mosquitoes, the pupation of caddisflies, and the disappearance of fairy shrimp, the invertebrate fauna becomes dominated by predatory species in late May. In addition to the species listed above, other migrants arrive, including giant water bugs (Belostomatidae), broad-shouldered water striders (Veliidae), water boatmen (Corixidae), water scorpions (Nepidae), and some additional beetle species. In addition, migratory green darner dragonflies (*Anax junius*) oviposit in large temporary ponds in April and May, and the voracious predatory larvae can become quite abundant in some ponds by mid-May. Phantom midge larvae (*Chaoborus*) also become abundant at this time. Many of these migrants are opportunistic and are not particularly adapted for ephemeral habitats, except that they are all characterized by rapid larval development.

There are few changes in the invertebrate faunal composition in June. With the advent of warm temperatures and less precipitation, the surface area and volume of these ponds begin to shrink, with concomitant increases in organism densities and nutrient concentrations. As the abundant predatory species increase their body sizes, shifts in their preferred prey may drastically alter relative abundances within the faunal assemblages. The general paucity of non-predatory macroinvertebrates at this time of the year means that predators are feeding on predators, and food webs may become very complex.

Adult *Sympetrum* dragonflies and *Lestes* damselflies begin emerging from the ponds by July 1, and *Anax* dragonflies emerge in mid- to late July from the larger ponds. By mid- July, most of the medium-sized ponds have dried, and the larger ponds have shrunk to only a small fraction of their maximum size. By this time, almost all insects have completed larval development and emerged as adults, and cladocerans have produced abundant ephippia, or drought-resistant eggs. The active invertebrate fauna at this time is characterized by adult insects (primarily bugs and beetles) capable of flying to permanent water, as well as other invertebrates that can burrow into the moist soil and/or form a drought-resistant stage (e.g., gastropods, planarians, and ostracods).

Even the largest of the temporary woodland ponds usually lose all surface water by early August. Undoubtedly, there are some insect larvae that do not complete development by this time and perish. In drought years, even insects that are well adapted to ephemeral habitats may become stranded. In most years, however, insects that perish from dessication are either typical temporary pond migrants (i.e., Group 4 of

Wiggins et al. 1980) that failed to complete development, or they represent oviposition mistakes by insects more typical of permanent water. An example of this latter category is the presence of early-instar dragonfly larvae of the genera *Libellula* and *Aeshna* in some of the larger temporary ponds during the summer. These insects are typical residents of permanent ponds and most species require at least one year for larval development. In years of high precipitation in which some of the usually temporary ponds do not dry, these insects may survive and complete development the following year. The usual consequence of such oviposition mistakes, however, is complete larval mortality (Higgins, unpubl. data).

SEASONAL PATTERNS—AESTIVAL PHASE

In most years, heavy precipitation during the mid to late summer can cause dry (or nearly dry) basins to flood again, triggering another cycle of invertebrate activity. The surface area of flooding during this aestival phase is generally less than half that of the much more extensive vernal phase. There are a few invertebrates that appear to be specifically adapted to this later period of flooding. The floodwater mosquitoes, *Aedes vexans* and *Aedes trivittatus*, as well as mosquitoes in the genus *Psorophora* are particularly well-adapted to summer rain pools. Although some eggs may hatch in the spring along with other species of *Aedes*, most *A. vexans* and *A. trivittatus* eggs, and all those of *Psorophora*, hatch following reflooding in the summer (Carpenter and LaCasse 1955). Unlike spring species of *Aedes* that oviposit primarily near the margins of the vernal extent of flooding, *A. vexans* also oviposits extensively in the interior portions of pond basins (Enfield and Pritchard 1977), a strategy that ensures hatching during summer flood events. Development is extremely rapid, with first-instars appearing within a few hours of flooding and adults emerging in less than a week. Densities of *A.*

vexans larvae can reach several hundred per liter in these habitats (Dixon and Brust 1972), and the large number of biting adult females that emerge make this species a serious pest of humans during the summer (Carpenter and LaCasse 1955; Wood et al. 1979). Another species of mosquito, *Psorophora ciliata*, that may have co-evolved with *A. vexans*, is predatory in larval instars II-IV, feeding primarily on *A. vexans* larvae (Breeland et al. 1961).

Other inhabitants of these aestival pools are either permanent residents (e.g., small crustaceans, planarians, gastropods), or opportunistic migrants (e.g., *Anopheles* mosquitoes, several species of beetles and bugs). This latter group includes adult insects of species typical of more permanent water, some of which may oviposit and attempt to complete an addtional generation in these summer rain pools. While some of these migrants appear within 1 or 2 days of flooding (e.g., *Anopheles* mosquitoes), predatory beetle larvae (e.g., *Acilius*), as well as most other predators, do not appear until several days after inundation. This lag time between inundation and the appearance of predators allows the rapidly-developing mosquito larvae to feed and grow relatively unmolested. Drought-resistant eggs that were deposited by insects and other arthropods that are well-adapted to temporary ponds do not hatch at this time because they require a cold period followed by a warm-up in order to break their diapause (Horsfall and Fowler 1961; Wiggins et al. 1980). In addition, most of these eggs are deposited near the margins of the vernal extent of flooding and are not inundated by summer flood events.

The aestival phase is usually very brief, with surface water persisting for only a month or less. Animals that are specifically adapted to aestival pools, such as the mosquito species listed above, must be capable of extremely rapid development for this life history strategy to be successful. This strategy can be viewed as an evolutionary tradeoff between risks and benefits. Although there is the risk of desiccation before larval development is completed, the larvae occupy a warm, nutrient-rich, and relatively predator-free environment in which development can occur rapidly.

TROPHIC RELATIONSHIPS

Temporary woodland ponds are detritus-based, heterotrophic habitats, with energy flow stemming predominantly from the leaf litter that falls into the basins. Emergent, submergent, and floating vascular plants are not common, and thus contribute little to the overall energy budget. Although primary production in the form of algal photosynthesis takes place, the intense shading by the surrounding woods in these ponds reduces its input compared with more open bodies of water, particularly later in the spring (Moore 1970). Algal production that occurs in the early spring prior to tree leaf-out, however, may provide a significant food source for filter-feeding organisms (e.g., cladocerans).

Leaf litter that falls into the dry basins in the autumn is initially colonized by terrestrial microbes (principally fungi) that begin the process of decomposition. Barlocher et al. (1978) examined protein and fungal biomass levels in experimental leaf packs placed in vernal pools in Ontario. Higher protein levels (corresponding to higher levels of fungal biomass) were observed in leaf packs that were exposed to terrestrial microbes and aerobic decomposition compared with leaf packs that were submerged in water for the same period of time. All protein levels declined rapidly, however, following submergence in the spring. The authors concluded that the protein-rich detritus of temporary ponds supports the required rapid development of animals that inhabit these ephemeral environments (Barlocher et al. 1978).

The rapid decline in leaf litter protein levels following submergence observed by these researchers suggests, however, that there is more than just high-protein detritus supporting temporary pond fauna through larval development. The relative paucity of shredding detritivores in these habitats (Figure 2), compared with many lotic situations, also suggests that other trophic pathways may be more important than direct feeding on leaf litter. Indeed, the early spring fauna is characterized by a diverse filter-feeding guild comprised of cladocerans, ostracods, fairy shrimp, and—at least part time—mosquito larvae. Larvae of *Aedes* mosquitoes, in addition to filtering micro-organisms and detritus from the water column, are also known to graze biofilm from the surfaces of leaf litter (Merritt et al. 1992). These larvae can apparently grow equally well filtering pond water alone as they can when provided with detritus on which to graze. In a field experiment conducted in 1996, we examined the growth of 30 first-instar mosquito larvae (*Aedes stimulans*) in each of 15 microcosms provisioned with either conditioned leaves, non-conditioned leaves, or no leaves. No difference in time to adult emergence or adult weight was observed among the three treatments (Table 2). Walker and Merritt (1988) reported similar results with the treehole mosquito, *Aedes triseriatus*). These results suggest an abundance of planktonic food sources of at least equal importance to the trophic hierarchy as the enriched leaf detritus. But are the two related?

It has been demonstrated that the degree of microbial colonization of leaf litter plays an important role in subsequent growth rates of invertebrate detritivores in both terrestrial and aquatic habitats (e.g., Barlocher et al. 1978; Suberkropp et al. 1983; Lawson et al. 1984; Merritt et al. 1984; Arsuffi and Suberkropp 1989; Walker et al. 1997). The dry phase of temporary ponds appears to be the most important period of

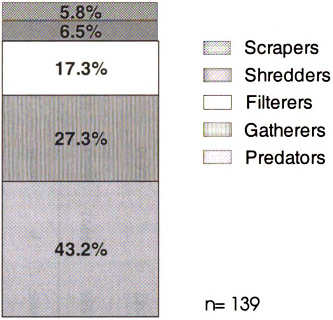

Figure 2. Functional feeding group composition (by genera) of invertebrates in temporary woodland ponds.

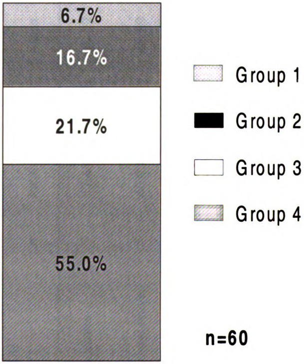

Figure 3. Predator composition by life history strategy in temporary woodland ponds.

Table 2. **Growth of first-instar *Aedes stimulans* mosquito larvae in field microcosms supplied with no leaves or with 3g (initial dry wt.) of conditioned or unconditioned leaves, Wild Ginger Pond, Lansing, Michigan 1996.**

		Days to Emergence				*Adult Weight (mg)*			
		MALES		*FEMALES*		*MALES*		*FEMALES*	
MICROCOSM TREATMENT	*n*	*mean*	*p*	*mean*	*p*	*mean*	*p*	*mean*	*p*
Conditioned leaves	5	47.43	0.9128	49.22	0.8758	0.583	0.9992	0.641	0.978
Non-conditioned leaves	5	46.93		48.89		0.587		0.636	
No leaves	5	47.41		49.39		0.586		0.662	

microbial colonization of leaf litter, due to the prevalence of terrestrial fungi as the major decomposers (Barlocher et al. 1978). This microbial colonization process begins as soon as water levels begin to decline in the late spring or summer. As a pond loses surface water and shrinks in size, the leached-out and highly-recalcitrant leaf detritus from the previous year becomes exposed and the moist surfaces are rapidly colonized by a host of fungi, bacteria, and protozoa. The celluloses and hemicelluloses contained in the leaves are principally exploited by terrestrial fungi, which depolymerize these complex compounds via cellulolytic enzymes into simpler carbohydrates (Ljungdahl and Eriksson 1985). Terrestrial fungi, principally white-rot fungi, are also important in breaking down lignin, which binds to the celluloses and hemicelluloses in leaves (Ljungdahl and Eriksson 1985). Although some lignin degradation occurs during the aquatic phase (Chamier 1985), the process is slow compared to that which occurs during the terrestrial phase, and much of the celluloses and hemicelluloses remain bound and unavailable for enzymatic degradation (Webster and Benfield 1986). The leaf surfaces are also colonized during this terrestrial phase by bacteria and their protozoan predators (Bamforth 1977), many of which secrete extracellular compounds (Nalewajko 1977). A similar colonization occurs on leaves that fall into the dry basins in the autumn. In this way, the complex structure of the leaves is slowly converted into microbial biomass.

It is suggested here that when the ponds reflood in the spring to their maximum size (and when they partially reflood in the summer to form aestival ponds), the heavily-enriched surfaces of the leaf detritus provide a nutrient broth in the form of dissolved and fine particulate organic matter stemming from the inundated fungal biomass, by-products of lignin degradation (Kirk 1984), partially degraded celluloses, and the biomass and extracellular compounds of bacteria and protozoa. These dissolved substances, principally dissolved organic carbons and nitrogens (DOC and DON), are used by planktonic and attached heterotrophic bacteria, and a microbial bloom ensues. At this same time, algal growth may also be stimulated from DON and the sunlight available before trees leaf out in the spring. The resulting growth in the microbial community, in turn, supports the vast filter-feeding guild characteristic of the early spring fauna of temporary woodland ponds. In addition, following inundation, aquatic hyphomycetes readily colonize and degrade the dead terrestrial fungal cells (which contain little or no lignin), resulting in a substantially higher biomass of hyphomycetes and associated micro-organisms than would be possible with the recalcitrant leaf detritus alone (Chamier 1985). This helps support invertebrate scrapers and gathering collectors, as well as shredding detritivores.

Trophic relationships within temporary woodland ponds are closely tied to the seasonal succession and adaptational strategies (Table 1) of the associated fauna. Animals with drought-resistant stages may begin/resume development soon after the ponds reflood, triggered by physio-chemical cues within the water(Horsfall 1956; Horsfall and Fowler 1961). The organisms that initially appear early in the spring are generally those that take advantage of the microbially-enriched water and detritus, i.e., filter-feeders, gathering collectors (e.g., certain chironomid midge larvae), and detritivorous shredders (primarily limnephilid caddisflies). These species can apparently tolerate the low temperatures and frequent ice cover characteristic of this time of the year. Even though growth is slower in this cold environment (Atkinson 1994), development occurs in a relatively nutrient-rich and predator-free environment. This strategy of beginning development early in the spring may have initially evolved as an adaptation for survival in small, ephemeral habitats. A reinforcing factor may have been predator avoidance in time. By hatching early, organisms such as mosquito larvae and fairy shrimp--animals with few defensive mechanisms--feed and grow relatively unmolested by predators. The majority of predators do not appear until weeks later. By the time most of the predatory larvae of the odonates and beetles make their appearance, these potential prey species are generally too large for the small, early-instar predators to effectively catch. Insects that develop in cold temperatures reach larger body size than those reared at higher temperatures (Brust 1967; Atkinson 1994), and the larvae of spring species of *Aedes* are some of the largest in this genus (Wood et al. 1979). Thus, the early spring inhabitants of temporary ponds may escape predation in time by beginning development early, and by achieving larger body sizes than their potential predators. As previously stated, only some dytiscid beetles in the genus *Agabus* that overwinter in the egg stage have apparently evolved to take advantage of the abundant prey available early in the spring. By beginning development early in the spring, larvae of these beetles are able to exploit even small ponds of only a few weeks duration, habitats unavailable for other beetle species that appear later in the season.

Although there are relatively few predators early in the spring, the number of predatory species increases substantially as the water temperature rises. Predator diversity reaches its peak in mid-May, with predators often becoming the dominant functional group within the ponds at this time. Most predators in temporary woodland ponds can be classified as either opportunistic migrants (Group 4 of Wiggins et al. 1980) or cyclic colonizers (Batzer and Wissinger 1996; Wissinger 1997), i.e., they generally lack specific adaptations to survive drought and overwinter in permanent-water habitats. Figure 3 illustrates that over half the predatory genera found in these ponds belong to this life history category. More striking perhaps is the very high proportion of predators within the Group 4 category itself (Figure 4). The dominance of predators in this category is not terribly surprising given that animals in this group must recolonize ponds every year, breed, and complete larval development before the ponds dry. The consumption of animal protein allows the larvae of these relatively late-arrivals to develop very quickly, the assimilation efficiency of predators being highest among trophic groups (Wotton 1994). Relatively rapid development is a characteristic of most animals breeding in ephemeral habitats, particularly those organisms that lack adaptations for coping with the dry phase.

Conversely, this probably explains the total lack of shredding detritivores exhibiting a Group 4 strategy (Figure 4). Although the leaf litter of temporary ponds may be enriched by the microbial processes described above, it is still a relatively poor food source, and detritivorous animals must consume large quantities to maintain growth. One of the few shredding insects, the caddisfly *Limnephilus indivisus*, begins development early in the spring--sometimes hatching the previous fall in autumnal ponds (Wiggins 1973)--a strategy which helps to ensure that the 8-10 week development will be completed before the pond dries. For a migrant species that arrives at a pond later in the spring, a feeding strategy based on consuming leaf detritus would be risky; growth would be slow and there would often be insufficient time to complete development. Interestingly, limnephilid caddisfly larvae in temporary ponds are known to be occasionally cannibalistic (Wissinger et al. 1996), and have been reported to be infrequent predators of mosquito larvae (Downe and West 1954; Baldwin et al. 1955). Cargill et al. (1985) demonstrated the importance of lipids in the diet of final-instar shredding caddisflies. The accumulation of triglyceride reserves during the last larval

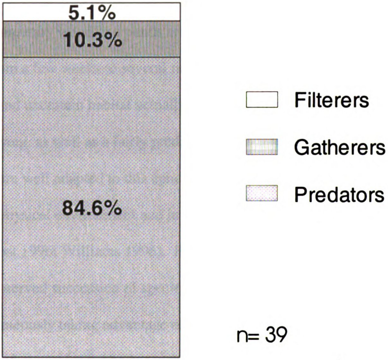

Figure 4. Functional feeding group composition for Wiggins et al. (1980) Group 4 life history strategy.

instar is apparently essential for completion of the larval stage and for subsequent adult reproduction (Cargill et al. 1985). Although lipids are available in aquatic hyphomycete fungi (Cargill et al. 1985), incidences of cannibalism and predation in otherwise detritivorous insects may be a means of quickly acquiring lipids as well as protein. This diet supplementation of protein and essential lipids may provide a boost in growth rates and ensure survival through the larval period (Wissinger et al. 1996).

CONCLUSIONS

In summary, temporary woodland ponds are small, microbially-driven wetlands that may be flooded from a few weeks to several months out of the year. What may initially appear as a harsh and uncertain habitat actually exhibits a reasonably predictable cycle of flooding and drying, as well as a fairly predictable seasonal succession of species. Invertebrates that are well adapted to this ephemeral habitat have evolved within the constraints of the physical environment and in response to community interactions (Schneider and Frost 1996; Williams 1996). Both abiotic and biotic factors appear to have shaped the observed succession of species, with early-season, cold-adapted inhabitants simultaneously taking advantage of the microbially-enriched environment and minimizing predation pressure by beginning development before most of the predators appear. Many of the insects that arrive later in the spring are migrants with no special adaptations to the dry phase of these ponds. The vast majority of these species are predatory in at least the larval stage, employing a trophic strategy that permits rapid growth in a shrinking environment.

REFERENCES

Arsuffi, T. L. and K. Suberkropp. 1989. Selective feeding by shredders on leaf-colonizing fungi: Comparison of macroinvertebrate taxa. Oecologia 79: 30-37.

Atkinson, D. 1994. Temperature and organism size--a biological law for ectotherms? Advances in Ecological research 25: 1-57.

Baldwin, W. F., H. G. James, and H. E. Welch. 1955. A study of mosquito larvae and pupae with a radio-active tracer. Canadian Entomologist 87: 350-356.

Bamforth, S. S. 1977. Litter and soils as freshwater ecosystems, p. 243-256. *In* [ed.], J.Jr.Cairns, Aquatic Microbial Communities. Garland. New York.

Barlocher, F., R. J. Mackay, and G. B. Wiggins. 1978. Detritus processing in a temporary vernal pool in southern Ontario. Archiv für Hydrobiologie 81: 269-295.

Batzer, D. P. and K. A. Sion 1999. Autumnal woodland ponds of western New York: Temporary habitats that support permanent water invertebrates, p. 319-332. *In* [eds.], D.P.Batzer, R.B.Rader, and S.A.Wissinger, Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley and Sons. New York.

Batzer, D. P. and S. A. Wissinger. 1996. Ecology of insect communities in nontidal wetlands. Annual Review of Entomology 41: 75-100.

Breeland, S. G., W. E. Snow, and E. Pickard. 1961. Mosquitoes of the Tennessee valley. Journal of the Tennessee Academy of Science 36: 249-319.

Brust, R. A. 1967. Weight and development time of different stadia of mosquitoes reared at various constant temperatures. Canadian Entomologist 99: 986-993.

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

Carpenter, S. J. and W. J. LaCasse. 1955. Mosquitoes of North America. University of California Press. Berkeley, CA.

Chamier, A. C. 1985. Cell-wall degrading enzymes of aquatic hyphomycetes: A review. Botanical Journal of the Linnean Society 91: 67-81.

Dixon, R. D. and R. A. Brust. 1972. Mosquitoes of Manitoba III: Ecology of larvae in the Winnipeg area. Canadian Entomologist 104: 961-968.

Downe, A. E. R. and A. S. West. 1954. Progress in the use of the precipitin test in entomological studies. Canadian Entomologist 86: 181-184.

Enfield, M. A. and G. Pritchard. 1977. Estimates of population size and survival of immature stages of four species of *Aedes* (Diptera: Culicidae) in a temporary pond. Canadian Entomologist 109: 1425-1434.

Horsfall, W. R. 1956. Eggs of floodwater mosquitoes (Diptera: Culicidae). III. Condition and hatching of *Aedes vexans*. Annals of the Entomological Society of America 49: 66-71.

Horsfall, W. R. and H. W. Jr. Fowler. 1961. Eggs of floodwater mosquitoes VIII. Effect of serial temperatures on conditioning of eggs of *Aedes stimulans* Walker (Diptera: Culicidae). Annals of the Entomological Society of America 54: 664-666.

Iversen, T. M. 1971. The ecology of a mosquito population (*Aedes communis*) in a temporary pool in a Danish beech wood. Archiv für Hydrobiologie 69: 309-332.

James, H. G. 1961. Some predators of *Aedes stimulans* (Walk.) and *Aedes trichurus* (Dyar) (Diptera: Culicidae) in woodland pools. Canadian Journal of Zoology - Journal Canadien de Zoologie 39: 533-540.

James, H. G. 1966. Location of univoltine Aedes eggs in woodland pool areas and experimental exposure to predators. Mosquito News 26: 59-63.

Kenk, R. The animal life of temporary and permanent ponds in southern Michigan. (71). 1949. University of Michigan, Museum of Zoology. Miscellaneous Publication.

Kirk, T. K. 1984. Degradation of lignin, p. 399-437. *In* [ed.], D.T.Gibson, Microbial Degradation of Organic Compounds. Marcel Dekker. New York.

Lawson, D. L., M. J. Klug, and R. W. Merritt. 1984. The influence of the physical, chemical, and microbiological characteristics of decomposing leaves on the growth of the detritivore *Tipula abdominalis* (Diptera: Tipulidae). Canadian Journal of Zoology - Journal Canadien de Zoologie 62: 2339-2343.

Ljungdahl, L. G. and K.-E. Eriksson. Ecology of microbial cellulose degradation. Marshall, K. C. (8), 237-299. 1985. New York, Plenum. Advances in Microbial Ecology.
Ref Type: Serial (Book,Monograph)

Merritt, R. W., R. H. Dadd, and E. D. Walker. 1992. Feeding behavior, natural food, and nutritional relationships of larval mosquitoes. Annual Review of Entomology 37: 349-376.

Merritt, R. W., W. Wuerthele, and D. L. Lawson. 1984. The effect of leaf conditioning on the timing of litter processing on a Michigan woodland floodplain. Canadian Journal of Zoology - Journal Canadien de Zoologie 62: 179-182.

Moore, W. G. 1970. Limnological studies of temporary ponds in south-eastern Louisiana. Southwestern Naturalist 15: 83-110.

Nalewajko, C. 1977. Extracellular release in freshwater algae and bacteria: Extracellular products of algae as a source of carbon for heterotrophs, p. 589-624. *In* [ed.], J.Jr.Cairns, Aquatic Microbial Communities. Garland. New York.

Schneider, D. W. 1999. Snowmelt ponds in Wisconsin: Influence of hydroperiod on invertebrate community structure, p. 299-318. *In* [eds.], D.P.Batzer, R.B.Rader, and S.A.Wissinger, Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley and Sons. New York.

Schneider, D. W. and T. M. Frost. 1996. Habitat duration and community structure in temporary ponds. Journal of the North American Benthological Society 15: 64-86.

Suberkropp, K., T. L. Arsuffi, and J. P. Anderson. 1983. Comparison of degradative ability, enzymatic activity, and palatability of aquatic hyphomycetes grown on leaf litter. Applied and Environmental Microbiology 46: 237-244.

Walker, E. D. 1995. Effect of low temperature on feeding rate of *Aedes stimulans* larvae and efficacy of *Bacillus thuringiensis* var. *israelensis* (H-14). Journal of the American Mosquito Control Association 11: 107-110.

Walker, E. D., M. G. Kaufman, M. P. Ayres, M. H. Riedel, and R. W. Merritt. 1997. Effects of variation in quality of leaf detritus on growth of the eastern tree-hole mosquito, Aedes triseriatus (Diptera: Culicidae). Canadian Journal of Zoology - Journal Canadien de Zoologie 75: 706-718.

Walker, E. D. and R. W. Merritt. 1988. The significance of leaf detritus to mosquito (Diptera: Culicidae) productivity from treeholes. Environmental Entomology 17: 200-206.

Webster, J. R. and E. F. Benfield. 1986. Vascular plant breakdown in freshwater ecosystems. Annual Review of Ecology and Systematics 17: 567-594.

Westwood, A. R., G. A. Surgeoner, and B. V. Helson. 1983. Survival of spring *Aedes* spp mosquito (Diptera: Culicidae) larvae in ice-covered pools. Canadian Entomologist 115: 195-197.

Wiggins, G. B. 1973. A contribution to the biology of caddisflies. Life Science Contributions of the Royal Ontario Museum 88: 1-28.

Wiggins, G. B., R. J. Mackay, and I. Smith. 1980. Evolutionary and ecological strategies of animals in annual temporary pools. Archiv für Hydrobiologie, Supplement 58: 97-206.

Williams, D. D. 1987. The Ecology of Temporary Waters. Croom Helm, Timber Press. Portland, OR.

Williams, D. D. 1996. Environmental constraints in temporary fresh water and their consequences for the insect fauna. Journal of the North American Benthological Society 15: 634-650.

Wissinger, S. A. 1997. Cyclic colonization in predictably ephemeral habitats: A template for biological control in annual crop systems. Bilogical Control 10: 4-15.

Wissinger, S. A., G. B. Sparks, G. L. Rouse, W. S. Brown, and H. Steltzer. 1996. Intraguild predation and cannibalism among larvae of detritivorous caddisflies in subalpine wetlands. Ecology 77: 2421-2430.

Wood, D. M., Dang, P. T., and Ellis, R. A. 1979. The mosquitoes of Canada (Diptera: Culicidae). The Insects and Arachnids of Canada, Part 6. Publication 1686. Ottawa, Ontario, Canada, Biosystematics Research Institute, Research Branch, Agriculture Canada.

Wotton, R. S. 1994. Particulate and dissolved organic matter as food, p. 235-288. *In* [ed.], R.S. Wotton, The Biology of Particles in Aquatic Systems. Lewis. Boca Raton, FL.

CHAPTER 2

FEEDING ECOLOGY OF *AEDES STIMULANS* (WALKER) MOSQUITO LARVAE IN TEMPORARY WOODLAND PONDS

ABSTRACT

The larval feeding ecology of a common vernal pool mosquito, *Aedes stimulans* (Walker) was studied using field microcosms and through quantitative analysis of larval gut samples. In addition, a comparison was made of the food of larvae from two very different vernal habitats, a small woodland vernal pool and a larger woodland pond. The microcosm experiments took place in the larger pond and were designed to examine the importance of grazing versus filtering for larval *A. stimulans* through the presence or absence of a grazeable substrate, testing for effects on survival, days to eclosion, and adult weight. No differences were observed, indicating that *A. stimulans* larvae in the larger pond were capable of obtaining adequate food from filtering the water column alone. Analysis of guts from the microcosm treatments indicated that larvae in the treatment with little grazeable substrate were consuming high quality flagellate phytoplankton from the water column. The ingestion of these algae was probably responsible for the sustained growth and survival of the mosquitoes. Analysis of both water and larval gut samples from the small vernal pool indicated that primary production is minimal in this habitat. Grazing microbially-colonized surfaces in this smaller, less-productive environment may be necessary due to the paucity of algae. Small vernal pools are probably more abundant on the landscape than larger ponds, and

grazing by *A. stimulans* larvae may be an adaptation for growth and survival in these less-productive and very ephemeral habitats.

INTRODUCTION

Temporary vernal pools are the larval habitats for many species of spring *Aedes* mosquitoes. In addition to being a nuisance to humans, the biting adults that emerge from these small pools and ponds are also potential vectors of disease, primarily LaCrosse and Jamestown Canyon encephalitis (Harwood and James 1979). The large numbers of mosquitoes produced by these small, ephemeral habitats (Enfield and Pritchard 1977) indicate that these ponds contain abundant larval food resources. Larvae of *Aedes* mosquitoes filter suspended material from the water column, but also spend considerable time grazing the biofilm from substrates (Horsfall 1955; Ameen and Iversen 1978; Merritt et al. 1992; Clements 1992). It has been generally assumed that larval grazing is a necessary component of mosquito feeding and growth (Fish and Carpenter 1982). Temporary woodland ponds contain abundant leaf detritus, and the microbial degradation of this leaf litter is believed to drive much of the energy flow within these systems (Barlocher et al. 1978). The role of leaf litter in the growth and survival of larval mosquitoes has been examined in detail for small container habitats such as treeholes (Fish and Carpenter 1982; Carpenter 1983; Walker and Merritt 1988; Walker et al. 1991; Merritt et al. 1992; Leonard and Juliano 1995; Walker et al. 1997; Aspbury and Juliano 1998), but has received relatively little attention with regard to other larval mosquito habitats (however, see Hinman 1930, Howland 1930). The role of leaf detritus in treehole ecosystems has proven to be complex. Leaves are colonized by microbes, which may then be grazed by mosquito larvae (Fish and Carpenter 1982), but leaves may also provide dissolved nutrients important for the growth of bacteria within the water column, which could be filtered by larvae (Walker and Merritt 1988; Walker et al. 1991; Merritt et al. 1992; Kaufman et al. 1999).

The objective of this research was to study the larval feeding ecology of a common vernal pool mosquito, *Aedes stimulans* (Walker), and examine the role of leaf detritus in survival, growth, and subsequent adult fitness. I hypothesized that larvae furnished with leaves on which to graze will exhibit increased survivorship, growth, and adult fitness compared to larvae without a suitable substrate on which to graze. I also compared the food of larvae from 2 very different vernal ponds.

MATERIALS AND METHODS

Study Sites

Feeding experiments were conducted at Wild Ginger #1 Pond (WG-1), which is a temporary woodland pond near Haslett, Michigan, USA (42° 45' 30" N, 84° 23' 50" W). This pond is approximately 600 m^2 when completely flooded, with a maximum depth of 0.8 m. In an average year, the pond begins to fill with water during the late autumn, and reaches its maximum size and depth in March from snowmelt and rainfall. The pond loses all surface water by mid July or early August. Water temperatures for 1996-1998 ranged from 4° C in late March to 25.5° C in mid June (Figure 1). The basin is situated within a 60-70 year-old deciduous woodlot of 70 ha and is surrounded by trees and shrubs, primarily red maple (*Acer rubrum*), American elm (*Ulmus americana*), black ash (*Fraxinus nigra*), swamp white oak (*Quercus bicolor*), and northern swamp dogwood (*Cornus racemosa*). The canopy does not completely close over the pond, but after the trees leaf out, most sunlight reaching the surface is indirect. Leaves from trees

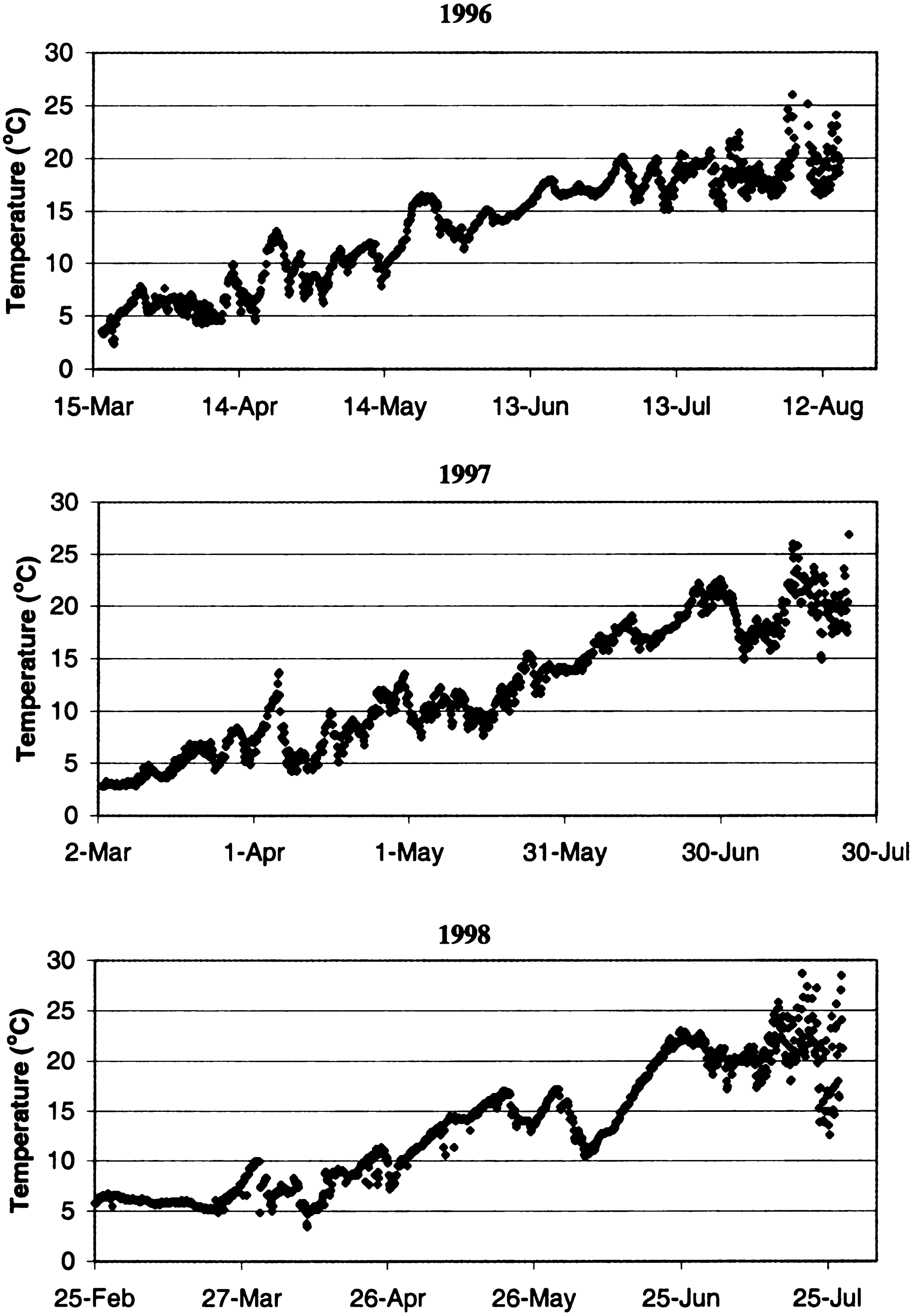

Figure 1. Water temperatures, Wild Ginger #1 Pond, 1996-1998.

near the basin and the surrounding upland (predominantly sugar maple and white oak) contribute to the extensive detritus that provides the energy source for the temporary pond community in the spring (Higgins and Merritt 1999). Wild Ginger #1 Pond supports a diverse invertebrate community, including large numbers of mosquito larvae in the early spring, with *Aedes stimulans* being the most abundant species.

Research was also conducted at a much smaller vernal pool, Hudson #1 Pond (HU-1), on the campus of Michigan State University, East Lansing, Michigan, USA. This pond covers an area of only 5 m^2 when flooded in the early spring, with a maximum depth of 0.5 m. From 1996-1999, the average duration of flooding was approximately 9 weeks, usually from late March through May. This small pool is situated in a mature beech-maple woodlot and, like WG-1, receives abundant leaf litter. The pond is completely shaded by the tree canopy when the trees leaf out in early May. In addition, leaf litter nearly fills the pond from top to bottom, further hindering light penetration. The invertebrate community of HU-1 is depauperate in comparison to WG-1, dominated by *Aedes stimulans* mosquito larvae and ostracods.

Microcosms

A field microcosm experiment to study the role of leaf litter in larval mosquito diets was conducted in WG-1 in 1996-1998. Microcosms were constructed from 4 l food-grade plastic containers measuring 17 cm in diameter at the top and tapering to 15 cm at the base (Figure 2). A hole 10 cm in diameter was cut in the base and two holes 10 x 10 cm were cut in the sides near the top. These holes were then covered with 250 µm Nytex nylon mesh. Curved Styrofoam pieces were glued between the upper holes and the top of the containers to keep the microcosms afloat (Figure 2). When placed in the

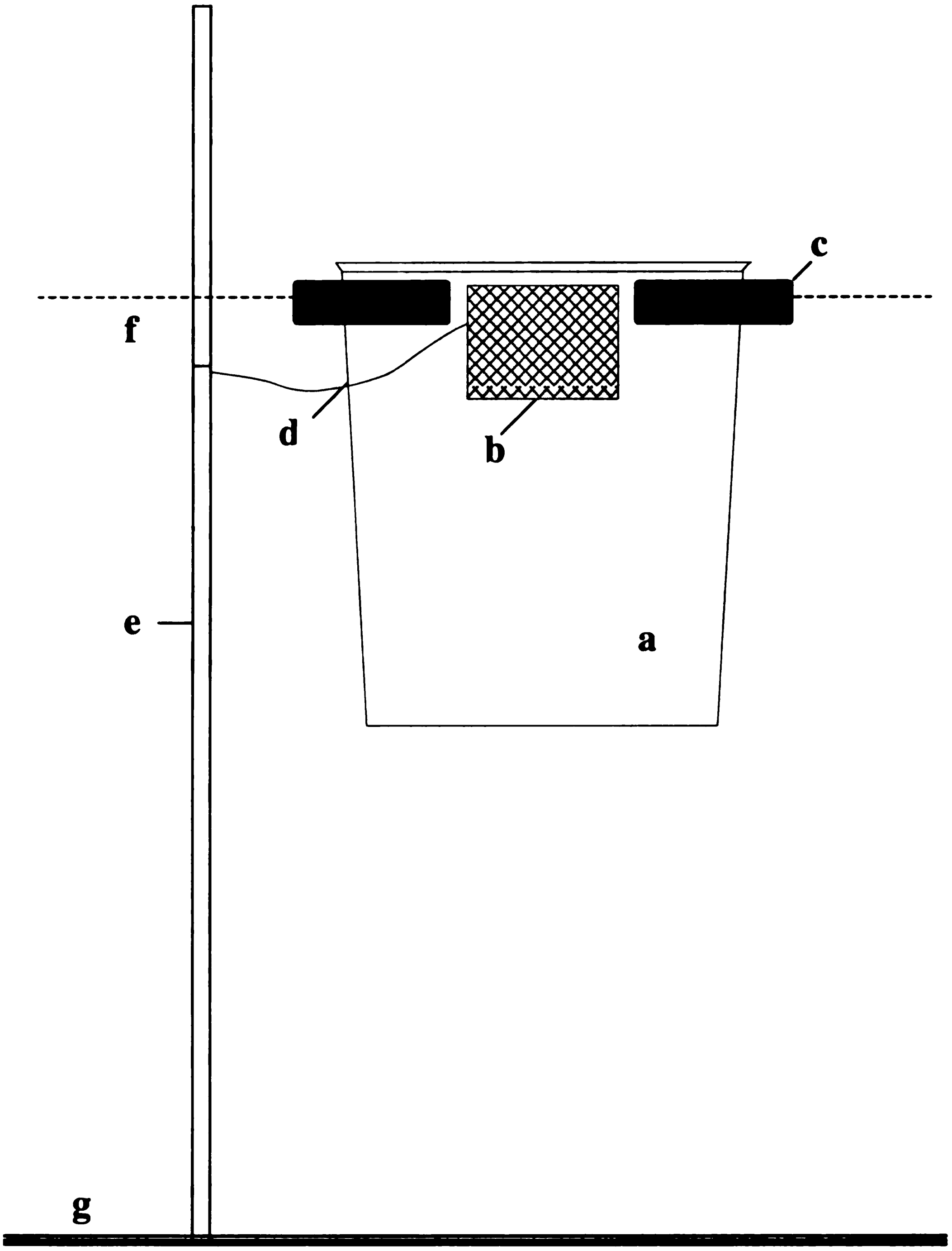

Figure 2. Diagram of floating microcosm used in field experiment. a) 4 l plastic container, b) 250 µm Nytex mesh on sides and bottom, c) Styrofoam floats, d) nylon fishing line to anchor microcosm, e) garden stake, f) water surface, g) substrate.

water the holes at the bottom and sides permitted pond water and most microbes to enter the microcosm, but excluded most invertebrates and kept mosquito larvae inside. Fifteen microcosms were placed into 60-70 cm deep water within WG-1 and anchored in place with garden stakes and fishing line. The microcosms were placed in groups of 3, representing the 3 treatments, to insure that each treatment within a group was exposed to similar micro-environmental conditions. In 1996, the treatments were 1) 3g of dried senescent sugar maple (*Acer saccharum*) leaves; 2) 3g of sugar maple leaves conditioned for 5 days in filtered (125 µm) pond water, and 3) no leaves. In 1997 and 1998, I wished to control for the potential build-up of biofilm inside the microcosms so the three treatments were: 1) 3g of dried leaves, 2) no leaves, with weekly replacement of the microcosm with a clean, dry one, and 3) no leaves and no microcosm replacement.

First instar (< 1 d old) *Aedes* mosquito larvae were collected from WG-1, and 30 were placed into each microcosm. It was not possible to make field identifications of mosquito larvae at that time, but all were subsequently identified (as adults or larvae) as *Aedes stimulans*. A plastic cover with 250 µm nylon mesh was placed over each microcosm to minimize the input of other detritus falling from the canopy above and to prevent predators from flying/crawling into the microcosms. When the microcosms were replaced each week in 1997 and 1998, larvae within the other treatments were also transferred briefly to another container and then returned to their original microcosm. This was done to minimize any effect of handling. When the larvae pupated, they were placed into small 0.5 l microcosms attached to the larger microcosms. These pupal containers were checked daily and adult mosquitoes, when present, were collected by microcosm and separated by date of emergence. Adults were immediately chilled in ice to minimize damage, and killed by freezing at -4° C. Each adult was subsequently identified to the species level, dried at 50° C for 48 h, and weighed to the nearest 0.0001g with a Cahn micro-balance.

Water and mosquito gut samples

Water column samples from WG-1 and HU-1 were collected on a monthly basis (early April and early May) for analysis of the microbial community. Water (2.5 ml) was collected with sterile pipettes, preserved immediately in 37% formaldehyde solution (final concentration 4%), and stored at 4° C. Five samples were collected each month. Samples were diluted 1:1 with deionized water and stained with the fluorescent dye 4'6-diamidino-2-phenylindole (DAPI) at a concentration of 30 μg/ml. In 1998, 5 fourth-instar larvae were collected from each treatment from one group of microcosms (designated a priori for such use), and 5 fourth instars were collected from HU-1. Following procedures outlined in Walker et al. (1988), the larvae were killed in hot water and preserved in 4% formaldehyde solution at 4° C. For each mosquito larva, the entire gut, with the perithrophic membrane intact, was removed with forceps and washed with deionized water. The contents of the gut was teased from the membrane with minuten pins in a drop of deionized water on a sterile microslide. The entire contents were pipetted into 2 ml of deionized water and shaken to dislodge particles. Because of the relatively large amount of material in the gut samples, only 1 ml of each sample (representing one-half gut) was processed, and subsequent counts were doubled. Using the procedures described by Hobbie et al. (1977) and Porter and Feig (1980), after staining with DAPI, gut and water samples were processed through a 0.22 μm black filter (backed by a 0.45 μm HA Millipore filter) using low vacuum pressure. The black filter was placed on a microslide that had been smeared with a thin layer of immersion oil. A drop of immersion oil was placed on the filter and a glass cover slip placed on top. All samples were stored in darkness at 4° C until counted. Algae were counted with an ocular grid at 250X using a Jenaluma microscope fitted for epifluorescence, a mercury lamp, and UV filter set. Bacteria were counted in a similar manner at 1000X, and identified as rods or cocci. Detritus particles < 10 μm were also counted at 1000X.

Statistical Analysis

Each microcosm was treated as the experimental unit. I tested for treatment effects on survival, days to adult emergence, and adult weight using a one-way analysis of variance (ANOVA) with the general linear models (GLM) procedure in SAS (SAS Institute 1990). Due to inherent differences between male and female mosquitoes in emergence and adult weight, sexes were analyzed separately. For survival analysis, sexes were combined, and proportions were analyzed using an arcsine transformation of the square root.

RESULTS

Microcosm Experiments

The results of the 1996 field experiment are shown in Figure 3. There were no differences in survivorship, days to adult emergence, or adult weight (Table 1). At the conclusion of this experiment, a thin biofilm was noted on the inside surfaces of the microcosms, including those with no leaves. Mosquito larvae certainly could have grazed on this biofilm, which could have been an adequate substitute for the biofilm on

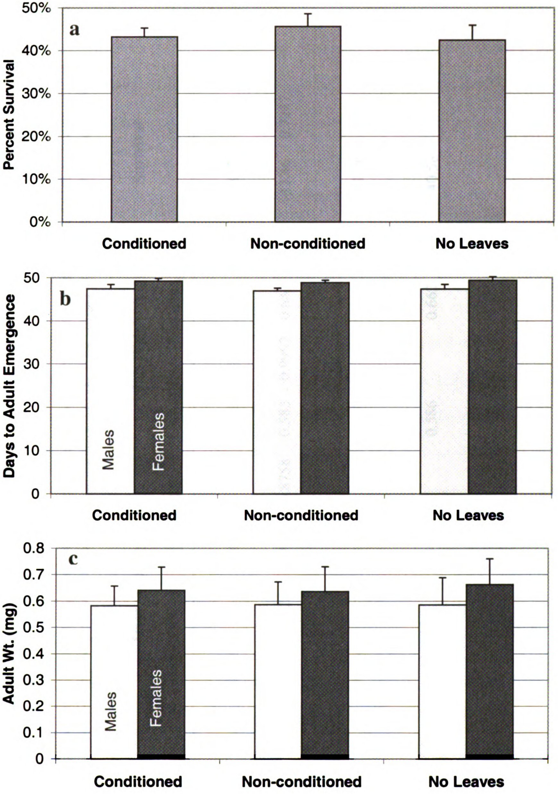

Figure 3. 1996 field microcosm results. a). survival, b). days to emergence, c). adult weight. Values are means and S.E.M.

Table 1. Growth of first-instar *Aedes stimulans* mosquito larvae in field microcosms supplied with no leaves or with 3g (initial dry wt.) of conditioned or unconditioned leaves, Wild Ginger Pond, Lansing, Michigan 1996.

		Days to Emergence				Adult Weight (mg)				Surivival	
		MALES		FEMALES		MALES		FEMALES			
MICROCOSM TREATMENT	n	mean	p	mean	p	mean	p	mean	p	mean	p
Conditioned	5	47.43	0.9128	49.22	0.8758	0.583	0.9992	0.641	0.9782	42.46	0.7237
Non-Conditioned	5	46.93		48.89		0.587		0.636		41.06	
No Leaves	5	47.41		49.39		0.586		0.662		40.58	

leaves in the other treatments. For this reason, the experimental design was altered the second and third years to include the weekly replacement of microcosms in one of the treatments with no leaves. While this did not totally eliminate microbial colonization of the microcosm surfaces, it did minimize it. It was believed that more frequent replacement would have subjected the larvae to increased handling stress. Also, because the dried leaves quickly became conditioned inside the microcosms, the conditioned-leaves treatment was dropped, keeping the number of treatments at 3.

The redesigned experiment was run in the spring of 1997. Unfortunately, a prolonged cold period during the first half of May (Figure 1) resulted in very heavy mortality of pupae in the microcosms. There was 100% mortality in 4 of 15 microcosms, and only 26 individuals survived to emerge as adults, with no microcosm yielding more than 4 adult mosquitoes. Mortality appeared to have been heaviest on females, perhaps due to their later pupation, with only 5 adult females emerging from the 15 microcosms. Six microcosms yielded only males and one treatment (no leaves-no microcosm replacement) yielded only 1 female. No analysis could be performed on adult weight or development time due to the small numbers of surviving adults. Analysis of survival data indicated that there were no differences in survival ($p = 0.8995$) among the 3 treatments.

In 1998, the experiment was repeated, again with weekly replacement of the microcosms in one of the treatments. Although mosquitoes in the replacement treatment were, on average, slightly smaller and took longer to emerge (Figure 4), these differences were not statistically significant (Table 2). Survival also was not significantly different among the 3 treatments.

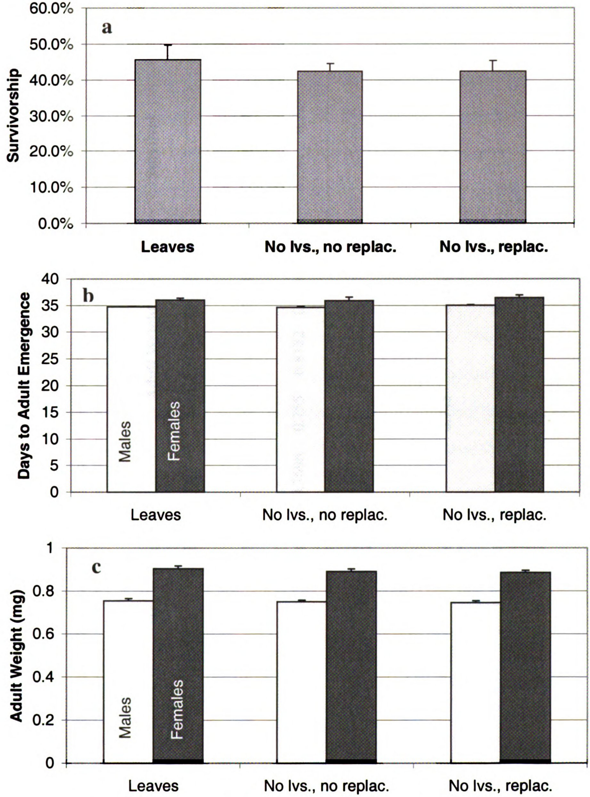

Figure 4. 1998 field microcosm results. a). survival, b). days to emergence, c). adult weight. Values are means and S.E.M.

Table 2. Growth of first-instar *Aedes stimulans* mosquito larvae in field microcosms supplied with leaves (3g initial dry wt.), no leaves, or with no leaves and weekly replacement of microcosms, Wild Ginger #1 Pond, Lansing, Michigan 1998.

		Days to Emergence				**Adult Weight (mg)**				**Surivival**	
		MALES		**FEMALES**		**MALES**		**FEMALES**			
MICROCOSM TREATMENT	**n**	**mean**	**p**	**mean**	**p**	**mean**	**p**	**mean**	**p**	**mean**	**p**
Leaves	5	34.74	0.1843	36.03	0.7696	0.755	0.8182	0.904	0.5309	42.44	0.7237
No Leaves, No Replacement	5	34.65		35.93		0.750		0.891		40.61	
No leaves, Replacement	5	35.05		36.43		0.745		0.886		40.59	

Water samples

Direct counts of algae, bacteria, and detritus for water samples from WG-1 and HU-1 are presented in Table 3. Protozoan numbers are not presented in this table as their numbers were surprisingly very low (< 5 ml^{-1}) in all samples. In addition, samples were not collected from HU-1 in 1996. While both ponds showed an increase and similar numbers of bacteria from April to May, the paucity of planktonic algae in HU-1 is notable. As previously stated, this pond contains water for only 8-10 weeks out of the year, and is heavily shaded by the forest canopy in May, conditions which may not be conducive for algal survival and growth. In addition, a large amount of leaf litter nearly fills the pond from top to bottom, further hindering light penetration. The dramatic increase in bacteria numbers from April to May in HU-1 may be attributed to the shrinking volume of the pond. The amount of algae in WG-1 remained fairly consistent from season to season, and in each year showed an increase in numbers from April to May.

One problem with comparing water samples by calendrical date among different seasons is that the phenology of these small bodies of water is largely dependent on hydrology (i.e., water volume) and temperature, which vary from year to year. For example, in 1996, WG-1 did not reach its maximum volume until the first week in April, and HU-1 was dry until that time. In 1998, both WG-1 and HU-1 reached their maximum extent of flooding the first week in March. In addition, there were differences in temperature between these two years (Figure 1).

Table 3. Direct counts of suspended material in water column samples using DAPI stain and epifluorescence microscopy. Values are cells or particles ml^{-1} (S.E.M.); n = 5 for each sampling date.

	Bacteria		Detritus	Algae
	Rods	Cocci		
Wild Ginger #1				
1996 April	1.83×10^{6}	3.56×10^{5}	1.08×10^{6}	1.04×10^{5}
	(1.02×10^{5})	(2.32×10^{4})	(1.11×10^{5})	(2.08×10^{4})
May	3.56×10^{6}	9.20×10^{5}	1.36×10^{6}	3.78×10^{5}
	(1.65×10^{5})	(3.73×10^{4})	(1.48×10^{5})	(1.89×10^{4})
1997 April	2.37×10^{6}	2.51×10^{5}	9.23×10^{5}	1.29×10^{5}
	(1.11×10^{5})	(1.39×10^{4})	(2.24×10^{4})	(1.62×10^{4})
May	3.06×10^{6}	7.47×10^{5}	1.19×10^{6}	3.11×10^{5}
	(2.18×10^{5})	(3.76×10^{4})	(1.26×10^{5})	(2.36×10^{4})
1998 April	2.79×10^{6}	4.49×10^{5}	1.25×10^{6}	1.71×10^{5}
	(2.12×10^{5})	(3.22×10^{4})	(2.03×10^{5})	(2.17×10^{4})
May	4.51×10^{6}	1.02×10^{6}	1.94×10^{6}	4.19×10^{5}
	(2.73×10^{5})	(8.44×10^{4})	(1.81×10^{5})	(2.97×10^{4})
Hudson #1				
1997 April	3.42×10^{6}	4.12×10^{5}	2.24×10^{6}	82
	(2.63×10^{5})	(3.69×10^{4})	(2.08×10^{5})	(21)
May	8.24×10^{6}	1.83×10^{6}	2.32×10^{6}	124
	(5.12×10^{5})	(8.47×10^{4})	(1.19×10^{5})	(37)
1998 April	2.89×10^{6}	5.74×10^{5}	1.97×10^{6}	108
	(1.32×10^{5})	(2.33×10^{4})	(1.48×10^{5})	(19)
May	9.16×10^{6}	1.69×10^{6}	2.78×10^{6}	132
	(4.05×10^{5})	(7.15×10^{4})	(1.76×10^{5})	(29)

Mosquito gut samples

Direct counts of food particles from larval gut samples are presented in Table 4. Of the larvae from the microcosms in WG-1, those from the treatment in which the microcosms were replaced showed distinct differences in the number and types of algae present in their guts. There were significantly fewer diatoms in these larvae compared to the other two treatments ($p < .001$), and more flagellate algae ($p < .05$). All larvae from each treatment contained abundant bacteria and detritus. The guts of larvae from HU-1 contained abundant detritus and bacteria, and, as expected, almost no algae (Table 4).

DISCUSSION

The hypothesis that grazing leaf litter is necessary for growth and survival of *Aedes stimulans* larvae, at least in WG-1 pond, can be rejected. That there were no differences in survivorship, days to eclosion, and adult weight among the 3 microcosm treatments in 1998 indicates that *A. stimulans* larvae are capable of growth by filter feeding alone. Larvae in the microcosm replacement treatment had very little material on which they could graze, yet performed equally well as those in the other 2 treatments. Water column samples from WG-1 indicate abundant bacteria, detritus, and algae (Table 3), and the gut contents of the microcosm-replacement larvae show that they were consuming large quantities of all of these (Table 4). In addition, the predominance of flagellate forms of algae in the guts of larvae in the replacement treatment suggests that these were obtained by filter feeding. *Chlamydomonas* and Cryptophyceae nannoplankton dominate the early season phytoplankton community in WG-1, and are important food sources for the zooplankton community (see Chapter 3). These are

Table 4. Direct counts of food particles from guts of fourth larval instars of *Aedes stimulans*. Values are means (n = 5) of cells or particles gut^{-1} and (S.E.M.).

	Wild Ginger #1 Microcosms			**Hudson #1**
	Leaves	**No Leaves, No Replacement**	**No Leaves, Replacement**	
Bacteria				
Rods	2.62×10^{6} (1.22×10^{5})	2.19×10^{6} (1.67×10^{5})	2.45×10^{6} (1.32×10^{5})	3.11×10^{6} (1.77×10^{5})
Cocci	4.71×10^{5} (2.29×10^{4})	3.92×10^{5} (3.01×10^{4})	4.41×10^{5} (2.78×10^{4})	6.27×10^{5} (4.23×10^{4})
Detritus	2.37×10^{6} (1.17×10^{5})	2.55×10^{6} (2.33×10^{5})	1.69×10^{6} (1.52×10^{5})	2.87×10^{6} (2.02×10^{5})
Algae				
Diatoms	430 (96)	314 (81)	26 (11)	0
Flagellates[a]	138 (31)	188 (34)	396 (103)	2.4 (0.75)
Other	20 (7)	32 (8)	152 (19)	0

[a] Flagellates primarily include *Chlamydomonas* and Cryptophyceae nannoplankton.

considered highly edible and nutritious forms of algae, even more so than diatoms (Schindler 1970; Porter 1973). The relatively large numbers of these flagellates in the guts of the replacement-treatment larvae may be the key to their survival and growth.

These data are similar to those found by Ameen and Iversen (1978), who examined gut contents and growth rates of *Aedes communis* and *A. cantans* from a temporary woodland pond in Denmark. Although only relative proportions are presented, and bacteria were not quantified in this study, gut contents of larvae were found to contain abundant *Chlamydomonas*. In laboratory growth experiments, larvae consuming only pond water grew at a somewhat slower rate than those provided with pond water and "bottom substrate" (Ameen and Iversen 1978). In the pond water only treatment, however, the water was changed only every second day, and the mosquito larvae removed 57% of the algal biomass during that time period (Ameen and Iversen 1978). Those short-term fluctuations in the food (algal) supply could have been responsible for the observed differences between the two treatments. In the present study, the screened openings in the microcosms allowed for a continuous influx of suspended material, including algae, which was representative of that found in the pond as a whole.

If larval *A. stimulans* mosquitoes are capable of obtaining sufficient food by filter feeding, why then do they graze? A clue may be provided by the data from HU-1. This very ephemeral and heavily shaded pond contained very little algae (Table 3), and gut samples of mosquito larvae from here are dominated by bacteria and detritus. In the microcosm experiments in WG-1, larvae with little material on which to graze were apparently able to receive adequate nutrition by ingesting suspended material, particularly high-quality algae. In HU-1, larvae did not have algae as a food source.

While they may be able to grow in such an environment by filter feeding bacteria and detritus from the water column, grazing the biofilm on leaf surfaces probably provides for faster growth in this very temporary habitat. The other invertebrate inhabitants of HU-1 include ostracods and a small population of amphipods, both essentially grazers. Small, shallow vernal pools like HU-1 are probably more abundant in the landscape than larger ponds like WG-1. *Aedes stimulans* is common to all of these vernal habitats, and grazing may be an adaptation for survival in the more common, but less productive shallow pools. Ideally, the microcosm experiment should be repeated in small ponds like HU-1, although the small, shallow nature of these pools would make such an experiment difficult.

The role of leaf detritus in temporary woodland ponds systems is complex. While there are a few detritivores that feed directly on leaves, the more indirect microbial route appears to be more important (Barlocher et al. 1978; Higgins and Merritt 1999). The release of dissolved organic carbon and nitrogen compounds from detritus drives benthic and planktonic microbial growth, which, in turn, provides the foundation for filtering and grazing guilds of invertebrates. In small ponds like HU-1 with little primary production, microbial colonization of leaves forms a biofilm that can be exploited by a relatively small group of grazing invertebrates. In larger ponds that receive sufficient light, the benthic grazing guild is accompanied by an extensive filter feeding guild, with mosquito larvae exploiting both benthic and planktonic environments.

REFERENCES

Ameen, M. and T. M. Iversen. 1978. Food of *Aedes* larvae (Diptera: Culicidae) in a temporary forest pool. Archiv für Hydrobiologie 83: 552-564.

Aspbury, A. S. and S. A. Juliano. 1998. Negative effects of habitat drying and prior exploitation on the detritus resource in an ephemeral aquatic habitat. Oecologia 115: 137-148.

Barlocher, F., R. J. Mackay, and G. B. Wiggins. 1978. Detritus processing in a temporary vernal pool in southern Ontario. Archiv für Hydrobiologie 81: 269-295.

Carpenter, S. R. 1983. Resource limitation of larval treehole mosquitoes subsisting on beech detritus. Ecology 64: 219-223.

Clements, A. N. 1992. The Biology of Mosquitoes. Chapman and Hall. London.

Enfield, M. A. and G. Pritchard. 1977. Estimates of population size and survival of immature stages of four species of *Aedes* (Diptera: Culicidae) in a temporary pond. Canadian Entomologist 109: 1425-1434.

Fish, D. and S. R. Carpenter. 1982. Leaf litter and larval mosquito dynamics in tree-hole ecosystems. Ecology 63: 283-288.

Harwood, R. F. and M. T. James. 1979. Entomology in Human and Animal Health, 7th. ed. Macmillan. New York.

Higgins, M. J. and R. W. Merritt 1999. Temporary woodland ponds in Michigan: Invertebrate seasonal patterns and trophic relationships, p. 279-297. *In* [eds.], D.P.Batzer, R.B.Rader, and S.A.Wissinger, Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley and Sons. New York.

Hinman, E. H. 1930. A study of the food of mosquito larvae (Culicidae). American Journal of Hygiene 12: 238-270.

Hobbie, J. E., R. J. Daley, and S. Jasper. 1977. Use of Nuclepore filters for counting bacteria by fluorescence microscopy. Applied and Environmental Microbiology 33: 1225-1228.

Horsfall, W. R. 1955. Mosquitoes: Their Bionomics and Relation to Disease. Ronald Press. New York.

Howland, L. J. 1930. Bionomical investigation of English mosquito larvae with special reference to their algal food. Journal of Ecology 18: 81-125.

Kaufman, M. G., E. D. Walker, T. W. Smith, R. W. Merritt, and M. J. Klug. 1999. Effects of larval mosquitoes (*Aedes triseriatus*) and stemflow on microbial community dynamics in container habitats. Applied and Environmental Microbiology [Appl.Environ.Microbiol.] 65: 2661-2673.

Leonard, P. M. and S. A. Juliano. 1995. Effect of leaf litter and density on fitness and population performance of the hole mosquito *Aedes triseriatus*. Ecological Entomology 20: 125-136.

Merritt, R. W., R. H. Dadd, and E. D. Walker. 1992. Feeding behavior, natural food, and nutritional relationships of larval mosquitoes. Annual Review of Entomology 37: 349-376.

Porter, K. G. 1973. Selective grazing and differential digestion of algae by zooplankton. Science 244: 179-180.

Porter, K. G. and Y. S. Feig. 1980. The use of DAPI for identifying and counting aquatic microflora. Limnology and Oceanography 25: 943-948.

SAS Institute, Inc. 1990. SAS/STAT User's Guide, Version 6, 4th ed. Cary, N.C.

Schindler, J. E. 1970. Food quality and zooplankton nutrition. Journal of Animal Ecology 40: 589-595.

Walker, E. D., M. G. Kaufman, M. P. Ayres, M. H. Riedel, and R. W. Merritt. 1997. Effects of variation in quality of leaf detritus on growth of the eastern tree-hole mosquito, *Aedes triseriatus* (Diptera: Culicidae). Canadian Journal of Zoology - Journal Canadien de Zoologie 75: 706-718.

Walker, E. D., D. L. Lawson, R. W. Merritt, W. T. Morgan, and M. J. Klug. 1991. Nutrient dynamics, bacterial populations, and mosquito productivity in tree hole ecosystems and microcosms. Ecology 72: 1529-1546.

Walker, E. D. and R. W. Merritt. 1988. The significance of leaf detritus to mosquito (Diptera: Culicidae) productivity from treeholes. Environmental Entomology 17: 200-206.

Walker, E. D., E. J. Olds, and R. W. Merritt. 1988. Gut content analysis of mosquito larvae (Diptera: Culicidae) using DAPI stain and epifluorescence microscopy. Journal of Medical Entomology 25: 551-554.

CHAPTER 3

CLADOCERAN SUCCESSION IN A TEMPORARY WOODLAND POND IN MICHIGAN: THE INFLUENCE OF PREDATION

ABSTRACT

Zooplankton and predator communities in a temporary woodland pond in southern Michigan, USA were sampled on a weekly basis from March through mid-July, 1999. Five species of cladocerans were recovered during this period. Early in the season, the only cladoceran present was *Daphnia ephemeralis*. This species underwent sexual reproduction and produced ephippial eggs in April, completing its life cycle for the season. Two forms of *Daphnia pulex* were evident at different times. Beginning in April, parthenogenetic *D. pulex* reached body sizes of 2.5-3.0 mm. This large early form underwent sexual reproduction in May, producing ephippial eggs, and thereafter parthenogenetic females of *D. pulex* rarely exceeded 1.6 mm in length. The later form also exhibited significantly longer tail spines relative to body size than the early form. Two other relatively abundant species, *Ceriodaphnia reticulata* and *Simocephalus exspinosus*, were found almost exclusively in the shallow portions of the pond, and exhibited relatively little season change in body sizes. Both seasonal succession and shifts in body size appear to be closely linked with shifts in the predator community. Early in the season, only small predators such as cyclopoid copepods and early-instar phantom midge (*Mochlonyx*) larvae are present. These predators would necessarily prefer smaller prey, such as juvenile cladocerans. The large adults of both *D. ephemeralis* and the early form of *D. pulex* were effectively immune to heavy predation

due to their body size. In May, larger predators such as larvae of the dytiscid beetle, *Acilius semisulcatus*, and larvae of *Ambystoma* salamanders appeared, preferring larger prey. Large body size for cladocerans then became disadvantageous. *Daphnia ephemeralis* had completed its life cycle by the end of April, and *D. pulex* began producing ephippia through sexual reproduction in early May. The smaller form *of D. pulex* that remained was apparently less susceptible to these larger predators due to a smaller body size, smaller size at first reproduction, and longer tail spine. *Simocephalus exspinosus* apparently reduced its exposure to predators by being more benthic than planktonic, attaching itself to detritus. *Ceriodaphnia reticulata*, which first appeared in the pond in early May, was probably too small to attract the larger predators present later in the season.

INTRODUCTION

Cladoceran community succession within lakes and permanent ponds has been examined extensively with regard to food resources and predation pressures. In contrast, the ecology of temporary pond zooplankton has received much less attention, particularly from a community perspective. Models of cladoceran succession based on data from permanent waters may be of limited utility in temporary habitats. Temporary ponds and pools are not small versions of their permanent counterparts, but have a hydrology, nutrient input, and fauna that are unique to this habitat (Wiggins et al. 1980; Williams 1987). Temporary ponds, for example, are not thermally stratified, and the shallow nature of these habitats precludes the existence of any aphotic zone. Small seasonal ponds located in woodlands generally receive much less direct sunlight than more open wetlands, and usually do not support emergent or floating macrophytes.

Invertebrate predators are numerically abundant, but salamander larvae (*Ambystoma* spp.) are often important predators of zooplankton. There are no fish in these seasonal ponds. The dry phase of temporary ponds is critical to the cycling of nutrients by increasing the rate of cellulose and lignin degradation in the leaf detritus compared to non-flooded or permanently-flooded habitats (Lockaby et al. 1996a; Lockaby et al. 1996b). The detrital pathways in these temporary habitats are very important, with microbial processes driving much of the nutrient cycling and energy flow within the system in both dry and aquatic phases of ponds (Higgins and Merritt 1999). The dry phase of these ponds also "resets" the system each year, making temporary woodland ponds ideal habitats for research.

MATERIALS AND METHODS

Study site

Research was conducted at a small woodland pool, Wild Ginger #1 (WG-1), near Haslett, Michigan, USA (42° 45' 30"N, 84° 23' 50"W). This pond is approximately 600 m^2 when completely flooded, with a maximum depth of 0.8 m. In an average year, the pond begins to fill with water during the late autumn, and reaches its maximum size and depth in March from snowmelt and rainfall. In 1999, the pond lost all surface water in mid July.

Water temperature in 1999 ranged from 5.5° C in late March to 25.5° C in mid June (Figure 1). The basin is situated within a 60-70 year-old deciduous woodlot of 70 ha and is surrounded by trees and shrubs, primarily red maple (*Acer rubrum*), American elm (*Ulmus americana*), black ash (*Fraxinus nigra*), swamp white oak (*Quercus bicolor*), and northern swamp dogwood (*Cornus racemosa*). The canopy does not

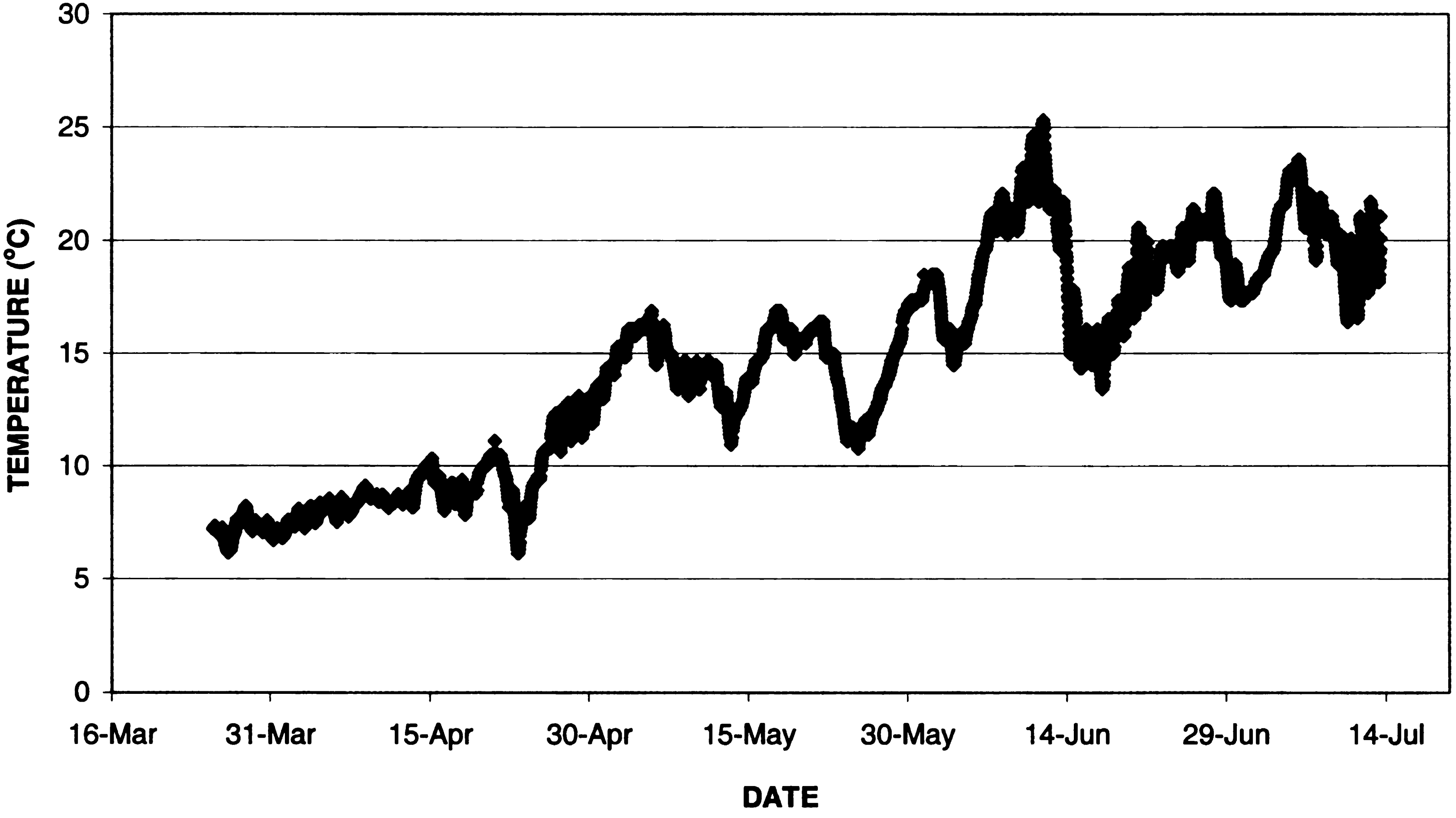

Figure 1. Water temperature, WG-1 Pond, 1999

completely close over the pond, but after the trees leaf out, most sunlight reaching the surface is indirect. Leaves from trees near the basin and the surrounding upland (predominantly sugar maple and white oak) contribute to the extensive detritus that provides the energy source for the temporary pond community in the spring (Higgins and Merritt 1999).

Collecting Methods

The zooplankton community was sampled on a weekly basis beginning when ice melted along the margins of the pond on March 15, 1999. Samples were collected by two different methods. A plankton net (mesh size 80 μm) was towed 3 times on each sampling date just below surface of the pond for a distance of 1.5 m. In the early spring, the net could not be towed due to ice cover. To sample near shore and during ice cover, a standard 500 ml mosquito dipper was used to collect water, which was then poured through the plankton net. This procedure was repeated 4 times for a total volume of 2 l at each of 3 sampling locations on each date. All samples were preserved in 70% ethanol and specimens were identified using keys in Pennak (Pennak 1989) and Wilson and Yeatman (1959). Cladoceran body lengths were measured to the nearest 0.03 mm using an ocular micrometer on a dissecting microscope. Body lengths for cladocerans do not include tail spines.

Larger invertebrates were collected in plankton tows at the same time the zooplankton was sampled. In addition to the invertebrates captured with the plankton net, the pond margins and interior were sampled with a D-frame aquatic net (mesh size 0.5 mm). The net was dragged for a distance of 1 m (total area 0.3 m^2) through the detritus near the shore, but only through the surface (0-20 cm) in the deeper part of the pond (in order to sample *Acilius* beetle larvae).

Water Samples

Water column samples from the pond were taken monthly for bacteria and algae counts. Water was collected with sterile pipettes, preserved immediately in 37% formaldehyde solution (final concentration 4%), and stored at 4° C. Five samples were collected each month. Samples were diluted 1:1 with deionized water and stained with the fluorescent dye 4'6-diamidino-2-phenylindole (DAPI) at a concentration of 30 µg/ml. Using the procedures described by Hobbie et al. (1977) and Porter and Feig (1980), each sample was processed through a 0.22 µm black filter (backed by a 0.45 µm HA Millipore filter) using low vacuum pressure. The black filter was placed on a microslide that had been smeared with a thin layer of immersion oil. A drop of immersion oil was placed on the filter and a glass cover slip placed on top. All samples were stored in darkness at 4° C until counted. Algae were counted at 250X using a Jenaluma microscope fitted for epifluorescence, a mercury lamp, and UV filter set. Algae was identified using reference keys (Prescott 1962; Prescott 1978; Cox 1996). Bacteria were counted in a similar manner at 1000X, and identified as rods, cocci, or spirilla. Detritus particles < 10 µm were also counted at 1000X.

RESULTS AND DISCUSSION

Seasonal Succession

Seasonal changes in species composition and density for the cladoceran community are shown in Figure 2. Increasing densities from late May through June probably reflect the decreasing size of the pond more than any increases in population sizes.

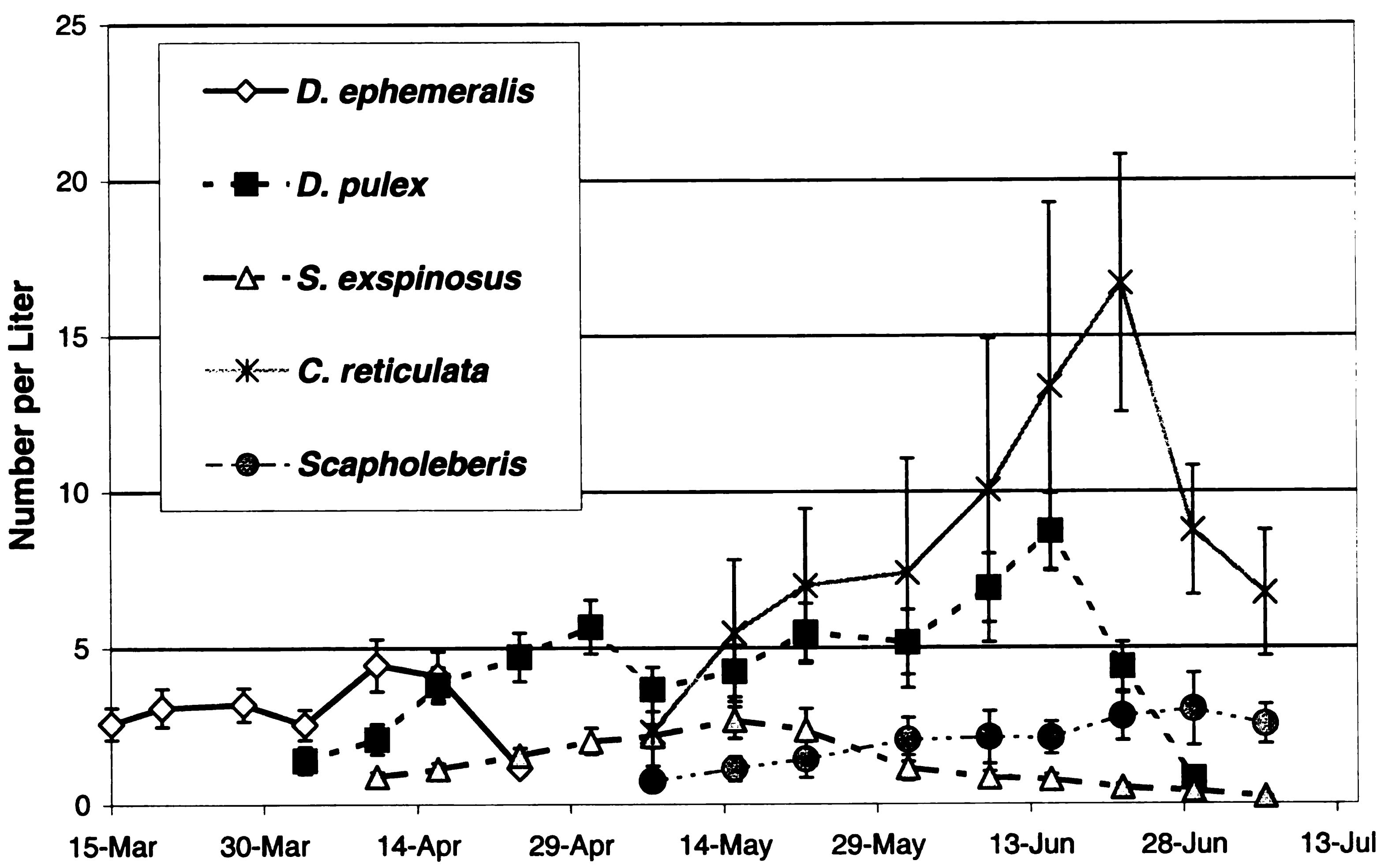

Figure 2. Seasonal changes in cladoceran density, WG-1 Pond, 1999.

Only 5 species were identified during the spring, and only 4 of these were abundant. Early in the spring, the only cladoceran present was *Daphnia ephemeralis* (Schwartz and Hebert). This species apparently is found only in temporary woodland ponds in the eastern United States (Schwartz and Hebert 1985). Adult parthenogenetic females were collected from beneath the ice in February, and were recovered along the margins of the pond as the ice melted in March. At that time the population was comprised primarily of parthenogenetic females, juvenile females, and a few males. By April 1, both males and sexually reproducing females became abundant, with ephippia being produced beginning shortly thereafter. No parthenogenetic females were collected after April 7, only males, unmated females, and ephippial females. *Daphnia ephemeralis* completed its life cycle by the end of April and no individuals were collected after April 24.

Daphnia pulex (Leydig) was first collected on April 3, shortly after *D. ephemeralis* began sexual reproduction, and about the same time that *Simocephalus exspinosus* (Koch) appeared in the pond. *D. pulex* became the numerically dominant cladoceran through the remainder of April and into May. Collections made May 7 through May 22 recovered numerous sexually reproducing females, males, and ephippial females of *D. pulex*. During this period, smaller parthenogenetic females of *D. pulex* appeared in the collections, exhibiting smaller body size and a longer tail spine than those recovered in April. This smaller form apparently replaced the earlier, larger form and was present until late June, shortly before the pond dried.

Ceriodaphnia reticulata (Jurine) was present in the pond from May 7 until the pond dried, and was the numerically dominant cladoceran after mid May. *C. reticulata* was primarily recovered in aggregations from the shallow margins of the pond, in association with *Scapholeberis mucronata* (O. F. Müller) and *Simocephalus exspinosus*. *Scapholeberis mucronata* was collected in very small numbers until late June (Figure 2). *Simocephalus exspinosus* was ubiquitous but never abundant, and was always found in association with detritus near the pond margins. The population of this species declined drastically during June as the pond shrank in size.

Seasonal Changes in Cladoceran Body Size

Body size measurements of parthenogenetic females for the four most abundant species in WG-1 are shown in Figure 3. Early in the spring, *Daphnia ephemeralis* females reached body lengths over 3mm. After the ice melted from the pond, these large individuals were only recovered from the interior portion of the pond, away from the margins. This distributional pattern probably reflects the warmer water temperature near shore (up to 7° C warmer) and the stenothermic cladoceran's avoidance of this warmer area. Body sizes of *D. ephemeralis* remained relatively large until the parthenogenetic individuals were replaced by smaller, sexually reproducing forms in early April. In April, *Daphnia pulex* females reached maximum sizes of 2.5-3.0mm, and showed a distribution pattern similar to the large *D. ephemeralis* individuals (i.e., occurring away from the margins of the pond). By May 7, however, the number of these large parthenogenetic females began to decline drastically, while the number of males and sexually-reproducing females rose even more drastically. During and after a period of intense ephippia production in May, parthenogenetic females of *D. pulex* collected from the pond were much smaller in size, rarely reaching body lengths over 1.6 mm. Only small parthenogenetic females of *D. pulex* were recovered through the remainder of the season. These smaller individuals also possessed significantly longer

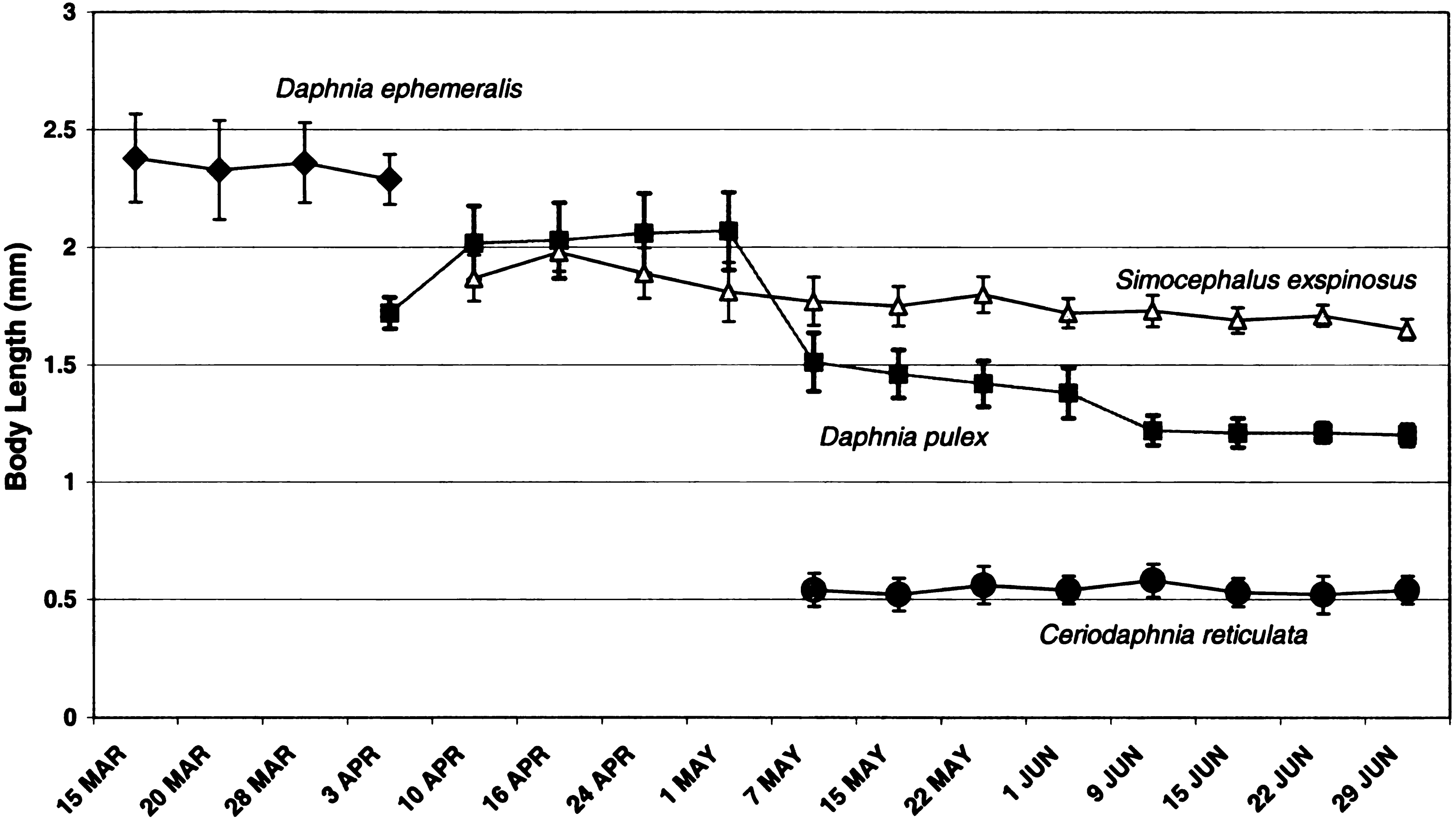

Figure 3. Seasonal changes in body lengths of parthenogenetic females of the four most abundant cladoceran species. Values are means (S.E.M.).

tail spines relative to body length than the *D. pulex* individuals collected in April. Body lengths of the other two abundant species, *Simocephalus exspinosus* and *Ceriodaphnia reticulata* showed less seasonal change than was exhibited by *D. pulex*, but *S. exspinosus* size decreased slightly as their numbers declined through June (Figure 3).

Predator Succession

Potential predators of cladocerans in WG-1 are listed in Table 1. Very early in the season few predators were present. In early March, the only predators collected were the cyclopoid copepods, *Diacyclops bicuspidatus thomasi* (Forbes) and the ubiquitous *Acanthocyclops vernalis* (Fischer). By mid-March, these were joined by the larger cyclopoid, *Macrocyclops albidus* (Jurine), and first and second instars of the phantom midge, *Mochlonyx cinctipes* (Coquillett). The 3 cyclopoid copepod species were not collected beyond April 24, but another, smaller cyclopoid, *Diacyclops navus* (Herrick) was present in May and June. *Mochlonyx* larvae were present through April. When *Aedes* mosquito larvae began hatching in substantial numbers in late March and early April, first instar larvae of the dytiscid beetle, *Agabus erichsoni* (Gemminger and Harold) also appeared in collections. Although this beetle apparently prefers mosquito larvae, it has been observed attacking cladocerans in a laboratory setting (Higgins, unpublished data). This species was most often found clinging to detritus near the pond's margins.

Odonate larvae of *Lestes* (primarily *dryas* Kirby) and *Sympetrum* (mostly *obtrusum* Hagen) began appearing in collections April 3. Teneral *Lestes* and *Sympetrum* adults were observed along the margins of the pond beginning June 15. First instar *Chaoborus americanus* (Johannsen) were collected on April 24, and third and fourth instars

Table 1. Major potential predators of cladocerans in WG-1 pond, 1999.

PREDATOR	**PERIOD PRESENT**	**DENSITY***
COPEPODA: CYCLOPOIDA		
Diacyclops bicuspidatus thomasi (Forbes)	March-April	1.3-2.4 l^{-1}
Acanthocyclops vernalis (Fischer)	March-April	0.6-1.8 l^{-1}
Macrocyclops albidis (Jurine)	March-April	0.09-0.12 l^{-1}
Diacyclops navus (Herrick)	May-June	1.1-1.8 l^{-1}
INSECTA: ODONATA		
Lestes dryas Kirby	April-mid June	0.7-2.6 m^{-2}
Sympetrum obtrusum Hagen	April-mid June	1.2-2.5 m^{-2}
INSECTA: COLEOPTERA		
Agabus erichsoni (Gemminger and Harold)	March-April	0.4-1.7 m^{-2}
Acilius semisulcatus Aubé	May	2.1 m^{-2}
INSECTA: DIPTERA		
Mochlonyx cinctipes (Coquillett)	March-April	0.9-2.7 l^{-1}
Chaoborus americanus (Johannsen)	late April-June	0.6-0.8 l^{-1}
AMPHIBIA: AMBYSTOMATIDAE		
Ambystoma sp.	May-June	0.07 m^{-2}

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*Densities listed as no. l^{-1} were determined using plankton tows. Densities listed as no. m^{-2} were determined using a D-frame net.

collected May 15. *Chaoborus* larvae, representing multiple generations, were present in the pond for the remainder of the season. A surprisingly significant predator of cladocerans in temporary woodland ponds is the dytiscid beetle larva, *Acilius semisulcatus* Aubé, which was first recovered on May 1 and was abundant through the entire month of May. Unlike many other dytiscid larvae that are found clinging to debris near the margins of ponds, first and second instars of *A. semisulcatus* were most often found near the surface in the deeper areas of the ponds. The larvae suspend themselves within the water column with their respiratory spiracles in contact with the surface and capture prey with their mandibles. Laboratory-reared first and second larval instars readily captured and consumed *Daphnia pulex* >2mm in length. Densities of *A. semisulcatus* first and second instar larvae within the interior portion of the pond (approximately 400m^2) on May 7 averaged 2.1 m^{-2} (range: 0-8 m^{-2}). *Acilius* larvae were found to be important predators of *Daphnia pulex* in a small pond near Montreal (Arts et al. 1981).

Larvae of salamanders (*Ambystoma* spp.) were present in relatively small numbers in WG-1 in 1999, although they were abundant in 1996 and 1997. Another temporary woodland pond 200 m to the west of WG-1 contained abundant salamander larvae in 1999, so the reason for their low numbers in WG-1 is unknown. Adult salamanders enter small woodland pools in late March or early April to breed and lay eggs, and the eggs hatch in 3-5 weeks, depending on temperature (Harding 1997). Larvae of *Ambystoma* are the only important vertebrate predators in small, temporary woodland ponds like WG-1, and they can have a significant impact on a zooplankton community as size-selective predators (Taylor et al. 1988).

Other Invertebrates

No discussion of the zooplankton community within a temporary pond would be complete without considering the impact of other filter-feeding organisms inhabiting the pond, principally *Aedes* mosquito larvae and the fairy shrimp, *Eubranchipus*. In 1999, the larvae of *Aedes* (primarily *stimulans* and *provocans*) began to hatch from eggs in late March (somewhat later than usual due to a dry winter), and reached their peak abundance after heavy rains in mid April. A larval density survey of the pond conducted on April 16 resulted in an estimate of 230,000-300,000 first and second instar mosquito larvae in the pond. Adult mosquitoes began to emerge in early May, and no larvae or pupae were observed later than May 15. Although *Aedes* larvae spend considerable time grazing the biofilm of leaf litter, they also filter feed with remarkable efficiency. Fourth instar *Aedes aegypti* larvae have been shown to clear water of yeast cells at the rate of 0.59-0.69 ml larva^{-1} h^{-1} (Aly 1988). Ameen and Iversen (1978) found that 9 third-instar *Aedes communis* larvae removed 57% of the algal biomass from 200 ml of pond water in 2 days. Gut contents of fourth-instar, filter-feeding *A. stimulans* larvae indicate an ability to filter particles ranging in size from bacteria (<1 µm) to large protozoa (> 100µm) (see Chapter 2).

Immature fairy shrimp (*Eubranchipus bundyi* Forbes) were already present in the pond when the ice began to melt in mid March. Growth was relatively fast; females carrying egg sacs were observed April 5, 10, and 16. By April 24 the population had declined drastically, and none were observed after that date. Density estimates for *E. bundyi* based on D-frame net samples were 15-20 individuals m^{-2} (9000-12,000 total individuals in the pond). *Eubranchipus bundyi* is a filter feeder, swimming ventral sideup as the legs move in a wave-like fashion, the legs serving as both means of locomotion and food-gathering device. Food particles are gathered through the beating motion of the legs into a ventral medial groove leading to the head. Food is reported to consist of algae, bacteria, protozoans, rotifers, and small detritus (Modlin 1982a; Pennak 1989).

Both *Aedes* mosquito larvae and fairy shrimp are common and usually very abundant members of the early spring fauna of temporary woodland ponds. Both are capable of filtering a wide range of particle sizes from the water column, and are potentially important competitors with the smaller zooplankton for algal resources. However, they may also play an important role in recycling nutrients. Crustacean zooplankton have been shown to be important in recycling nitrogen and phosphorus back into the water column where the nutrients can be reabsorbed by phytoplankton (Lehman 1980). Mosquito larvae in particular, through their grazing of leaf litter and production of feces, may provide an important link from benthic to planktonic food webs. Also, due to the mosquitoes' ability to filter and assimilate bacteria, an inadequate food source for cladocerans (Porter 1984), they may provide a link between planktonic bacteria and algae. Nitrogen and phosphorus contained within the feces could stimulate algal growth when released into the water, which would increase food availability to the zooplankton community. Clearly, additional research needs to be conducted regarding the competitive versus recycling relationship of large filter-feeding invertebrates with zooplankton in aquatic habitats.

In addition to these relatively large filtering-gatherers, the herbivorous calanoid copepod, *Aglaodiaptomus leptopus* (Forbes) was very abundant from mid-May through

June. By late May, the density of all life stages of this copepod reached 23 l^{-1}. The planktonic rotifer, *Hexarthra*, was common in May through June. This herbivorous, soft-bodied rotifer is relatively large at approximately 250 µm in length. By mid June, when most females were carrying resting eggs, the density of rotifers reached 37 l^{-1}. Other common filter-feeding organisms include the benthic fingernail clam, *Musculium*.

Seasonal Changes in the Microbial Community

Densities of bacteria, algae, and detritus are presented in Figure 4. Both bacteria and algae numbers increased from March through May, and then leveled off in June. Of particular interest to this study is the algal community, and a more detailed analysis of seasonal changes is presented in Figure 5. In March and April, the flagellates, *Chlamydomonas* and species of Cryptophyceae dominated the planktonic algae. By May, other green algae, particularly *Ankistrodesmus falcatus* and *Scenedesmus* (*quadricauda* and *opoliensis*) increased in number, while *Chlamydomonas* numbers remained relatively constant. As the pond began to shrink rapidly during June--with a concomitant rise in temperature--blue-green algae (principally filamentous forms such as *Oscillatoria*, *Anabaena*, and *Spirulina*) increased drastically and *Chlamydomonas* numbers dropped. The increase in the numbers of desmids and dinoflagellates late in the season may be attributable to grazing effects. Diatom numbers remained relatively constant through May, but were never abundant in the phytoplankton. Diatoms were much more abundant on the leaf litter in the pond (Higgins, unpubl. data). The most common diatom genera recovered from the water column were *Nitzchia*, *Navicula*, and *Gomphonema*.

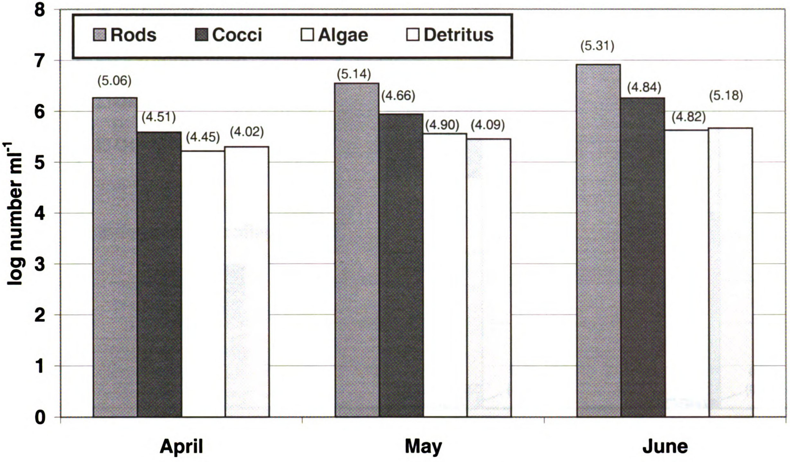

Figure 4. Microbial and detritus counts for water column samples, WG-1 Pond, 1999. Values are means (S.E.M.), n=5. All numbers are log transformed.

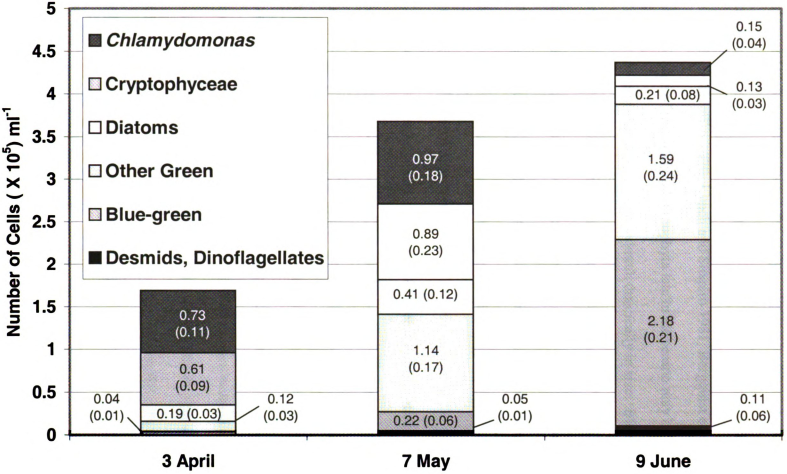

Figure 5. Seasonal changes in the algal community of the water column, WG-1 Pond, 1999. Values are means (S.E.M.), n=5.

Both *Chlamydomonas* and the Cryptophyceae are considered highly edible forms of algae, and their predominance in the early spring indicates a high-quality food source for cladocerans. The two common green algae, *Ankistrodesmus* and *Scenedesmus* are also edible (Schindler 1970; Lynch and Shapiro 1981; Bergquist et al. 1985). The predominance of filamentous Cyanophyta late in the season may be attributable to increases in temperature as the pond shrank drastically in size. Blue-green algae tend to predominate over other algal forms at temperatures above 20° C (Tilman et al. 1986). Blue-green algae, especially filaments, are considered poor food sources for cladocerans (Porter and Orcutt, Jr. 1980). Mats of filamentous algae never appeared, probably because of the limited amount of sunlight reaching the pond.

Successional Patterns and Predation

Successional and body size changes in cladoceran communities have been linked to both shifts in available food (algal) resources and predation pressure (Brooks and Dodson 1965; Hall et al. 1976; Zaret 1980; Romanovsky and Feniova 1985; Sommer et al. 1986; Gliwicz and Pijanowska 1989). Both food limitation and predation pressure can produce similar effects on the zooplankton community and it is often difficult to separate these two not necessarily mutually exclusive processes. In general, smaller-bodied cladocerans are better competitors in an environment with a fluctuating food supply (Gliwicz 1990). Although qualitative seasonal changes in the microbial community occurred, there is no indication that edible forms of algae declined until June, when blue-green algae began to predominate (Figure 5). Figure 4 shows that the quantity of algae and bacteria actually increased through the spring. In addition, by early May, the large filter-feeding organisms such as fairy shrimp and mosquito larvae

had completed their life cycle or exited the pond as adults. If food was the driving factor behind the observed changes in the cladoceran community, the departure of these larger competitors would be expected to lead to an increase in larger-sized cladocerans, rather than a decline (keeping in mind, however, that these larger invertebrates may be important in nutrient recycling). Finally, all cladoceran species did not exhibit shifts in body size at the same time. *Simocephalus exspinosus* body size, for example, remained relatively constant until June (Figure 3).

Much of the seasonal changes may be explained by shifts in predation pressure. Early in the spring, the predator community was comprised of copepods and early-instar *Mochlonyx* larvae. These early season predators are all selective toward small-sized prey. Brambilla (1982) found that 95% of *Daphnia pulex* consumed by third and fourth instar *Mochlonyx* larvae were < 0.80 mm long, so first and second instar *Mochlonyx* could be expected to prefer even smaller prey. Brandl and Fernando (1974) reported that *Acanthocyclops vernalis* preferred *Ceriodaphnia* prey < 0.32 mm long over those 0.40-0.54 mm in length. *Macrocyclops albidus*, being somewhat larger (up to 2.5 mm long), is capable of capturing larger prey (including first instar mosquito larvae), but appears to prefer prey < 1 mm long (Fryer 1957). Given the preference for small prey for these early predators, predation pressure on the only cladoceran present at that time, *Daphnia ephemeralis*, would have been heaviest on juveniles and smaller adults. After individuals reached a certain size, they were effectively immune to predation because virtually no large predators were present. The only early-season large predator was the dytiscid larva, *Agabus erichsoni*, which is capable of capturing large cladocerans. However, this beetle appears to prefer mosquito larvae and was recovered exclusively from debris near the margins of the pond, an area where large *D. ephemeralis* rarely occurred, probably due to the warmer water temperature.

The influence of predation on the seasonal patterns of *D. ephemeralis* also can be inferred by examining the defensive morphologies exhibited by this cladoceran. Neonates exhibit both a tail spine and a neck spine (Schwartz and Hebert 1985), morphological traits that reduce susceptibility to invertebrate predation (Pijanowska 1990; Repka et al. 1995b; Repka and Pihlajamaa 1996). Males and ephippial females, which are much smaller than parthenogenetic females (1.0-1.4mm), also possess a distinct tail spine (Schwartz and Hebert 1985). Adult parthenogenetic females of *D. ephemeralis*, however, show little in the way of defensive morphology; they even lack the tail spine that is characteristic of other members of the genus *Daphnia*. In their environment of small predators, *D. ephemeralis* clearly concentrates defensive strategies in smaller individuals.

One apparent drawback to large body size in cladocerans is an increase in vulnerability to egg predation by the copepodite stage of cyclopoid and calanoid copepods. These immature copepods are capable of entering the brood chambers of large (> 2.5 mm) cladocerans and feeding on the developing eggs (Gliwicz and Stibor 1993). Vulnerability to egg predation is apparently reduced by the presence of a large second abdominal process (Hanazato and Dodson 1995). Although egg predation in *D. ephemeralis* was not examined in this study, adult parthenogenetic females do possess a well-developed second abdominal process (see Schwartz and Hebert 1985), possibly a defense against egg predation by immature copepods.

What, then, is the reason for the relatively early departure of *D. ephemeralis* from the pond? Food limitations seem unlikely given that algal biomass is increasing during April (Figure 4). Food quality does not appear to have decreased (Figure 5), and the similarly-sized *Daphnia pulex* that replaced this species was able to grow and reproduce well on the available resources. Direct competition with *D. pulex* also seems unlikely given that sexual reproduction began before *D. pulex* appeared in the pond. The relatively early production of ephippia and the completion of the life cycle of *D. ephemeralis* in April may be viewed as an adaptive strategy to avoid heavy predation later in the spring. Ephippia production is completed prior to the appearance of such large predators as *Acilius* and *Ambystoma* larvae. With little in the way of defensive morphology to protect them, adult *D. ephemeralis* would certainly be vulnerable to heavy predation pressure. Although early-season ephippia production may have evolved as a consequence of the stenothermic nature of this species, as well as the ephemeral nature of the habitat, the advantage of predator avoidance has probably contributed to the maintenance of such a strategy. In this situation, the evolution of any trait extending the life cycle of this species would be selected against. Individuals with a higher temperature threshold, for example, would persist in the pond further into the season and be more likely to fall prey to the larger predators present at that time, thus reducing their fitness. In addition to avoiding predation, the early ephippia production by *D. ephemeralis* avoided any possible competition with the similarly-sized *D. pulex* that followed.

In terms of body size and cyclomorphosis, *Daphnia pulex* exhibited the most seasonal variation of any cladoceran species present in the pond. The pattern of large-sized individuals early, and then replacement by smaller individuals later in the spring has been described previously for temporary ponds (Brambilla 1980; Modlin 1982b) (Dodson 1974; Crosetti and Margaritora 1987) and permanent bodies of water (Hall 1964; Lynch 1978; Threlkeld 1979). Larger bodied cladocerans are able to feed more efficiently at lower food levels than smaller cladocerans (Gliwicz 1990), but are more vulnerable to size-selective predators (Brooks and Dodson 1965). The two forms from WG-1 differ in more ways than body size. Of particular interest are the presence of neck teeth in first instars of the large clone (absent in the small clone) and the longer tail spine on adults of the smaller form. Although cyclomorphosis in *D. pulex* is less pronounced than in some other *Daphnia* species, there can be considerable variation in the development of neck and tail spines (Black and Dodson 1990; Repka and Pihlajamaa 1996). In addition to these morphological differences, the late form begins reproduction at a smaller body size than the earlier form (0.9 mm versus 1.4 mm, respectively).

The larger-sized *D. pulex* compares favorably with *D. ephemeralis* in terms of defensive morphology. The presence of a neck tooth and a relatively long tail spine on first and second instars of *D. pulex*, and the short tail spines of large adults indicate that—as is the case with *D. ephemeralis*—predator defense is concentrated in smaller individuals. Through April, *D. pulex* shared the same predator species as *D. ephemeralis*, primarily cyclopoid copepods and *Mochlonyx* larvae (third and fourth instars during April). These predators would select smaller prey, and for *D. pulex*, this means early instars would be most vulnerable. With only these predators as the primary threat, larger individuals were at an advantage and could essentially escape predation.

This situation changed rather abruptly in early May, however. Predators like *Acilius* and *Ambystoma* larvae appeared. Unlike the early-season predators that selected for small individuals, these larger predators preferred larger prey. *Ambystoma* larvae apparently prefer prey $\geq$ 1.3 mm in length (Brambilla 1980). While no prey size preference has been determined for first and second instar *Acilius* larvae, individuals in a preliminary laboratory experiment captured and consumed *D. pulex* 1.8-2.5 mm long. For the early-season *D. pulex*, large size quickly became a disadvantage. At the same time these predators began to appear in the pond, *D. pulex* began to produce males, sexually-reproducing females, and—soon after—abundant ephippia. Thereafter, only smaller parthenogenetic females were recovered from the pond.

If the two forms of *D. pulex* in WG-1 represent different genotypes (which has yet to be determined), the production of ephippia by the larger clone in May appears to be a strategy to avoid predation. Predator avoidance through diapause is not unprecedented. Ślusarczyk (1995) found that the exudates of predatory fish triggered ephippia production in *Daphnia magna*. The production of diapausing eggs in the copepod, *Diaptomus sanguineus* (Forbes) has been shown to be heavily influenced by increased predation levels (Hairston and Munns 1984; Hairston 1987). At present, it is unknown whether the cues that triggered sexual reproduction in *D. pulex* in May came directly from the presence of predators or were based on an indirect environmental cue such as temperature or photoperiod. If temperature or photoperiod is found to be the proximate cause for ephippia production, this does not necessarily diminish the adaptive significance of entering diapause at a time in the season when predation pressure becomes severe. Predator-induced ephippia production can be viewed as a "bethedging" strategy (Schaffer 1974). Predation pressure may become severe enough to eliminate a population, and the production of resting eggs insures survival of the population into the following season. A pond 200 m west of WG-1 contained abundant *Ambystoma* larvae, and no *D. pulex* were recovered after May 22, suggesting that the salamanders eliminated this population.

The smaller, late season *D. pulex* exhibited different defensive morphology than the early season form. Neonates did not have neck a neck tooth, which was somewhat surprising given the presence of *Chaoborus* and copepod predators that would favor smaller prey. The presence of *Chaoborus* is known to induce the development of neck teeth in *D. pulex* (Schwartz 1991; Parejko 1991; Repka et al. 1994; Luning 1995), but the density of this predator in WG-1 may have been too low (< 1 liter^{-1}) to trigger this response. Another indication that predation pressure had shifted toward larger individuals of *D. pulex* can be seen in the development of the tail spine. In the early spring, tail spine length declined with increased body size (Figure 6), implying that larger individuals were less susceptible to predation. Later in the spring, tail spine length was positively correlated with body size (Figure 6), suggesting that larger individuals were now more vulnerable.

If the early, large-sized *D. pulex* is entering diapause as a means of avoiding predation, is the smaller form that follows less susceptible to these same predators? In preliminary feeding experiments with *Acilius* larvae, first instars attacked any prey over 1 mm long. Second instars showed little interest in *D. pulex* < 1.5 mm long, but readily attacked individuals > 2.0 mm long. Most of the late-season *D. pulex* would fall below the preferred prey size ≥ 1.3 mm for *Ambystoma.* The size at first reproduction

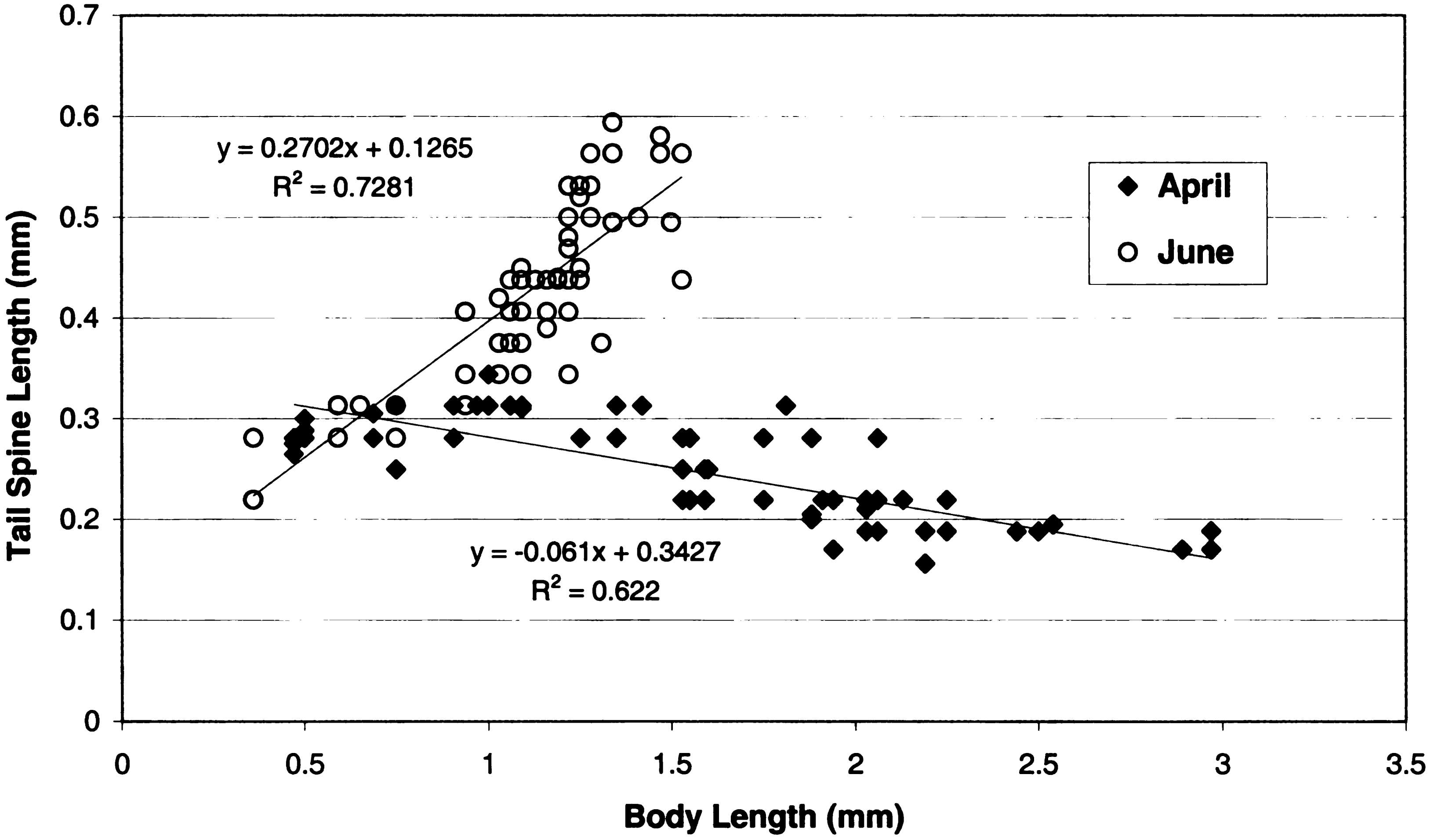

Figure 6. Tail spine length as a funtion of body length for parthenogenetic Daphnia pulex during early (April) and late (June) season. Slopes differ significantly at $p < 0.0001$.

for the late-season form is 0.9 mm, well below the preferred prey size of both *Ambystoma* and *Acilius* larvae. At the other end of the predator size spectrum, the two most important potential predators on smaller individuals are *Chaoborus americanus* larvae and the cyclopoid copepod, *Diacyclops navus*. Somewhat surprisingly, *Chaoborus* was found not to be an important predator of *D. pulex* in WG-1. An examination of 20 guts of *C. americanus* third and fourth instars found only 2 *D. pulex* remains. Calanoid copepods (probably *Aglaodiaptomus leptopus*) were most common, followed by soft-bodied rotifers (*Hexarthra*), and *Ceriodapnia reticulata*. Moore (1988) found rotifers to be the most abundant prey in all instars of *C. punctipennis*, and Brambilla (1982) also found *Chaoborus* larvae to be insignificant predators on *D. pulex* in a temporary pond near Ann Arbor, MI. The prey preferences of *D. navus* are not known for the field, but Brambilla (1982) observed that *D. navus* readily consumed first and second instar *D. pulex* in the laboratory. Therefore, it can be assumed that this copepod, although never abundant in WG-1, would prey on early instar *D. pulex* in the field.

The other two cladoceran species present in WG-1, *Ceriodaphnia reticulata* and *Simocephalus exspinosus*, showed relatively little size variation through the season compared to *D. pulex*. Both of these species were found primarily in shallow water along the pond margin, with *S. exspinosus* capable of withstanding the higher temperatures of this area of the pond (LaBerge and Hann 1990). *Simocephalus exspinosus*, like other members of this genus, spends much of its time attached to plant material by a sticky mucus on the dorsum of its carapace. In WG-1, individuals attached themselves to detritus on the bottom of the pond, making this species more benthic than planktonic. The predators that occupy this habitat—odonate and beetle larvae—are sit-and-wait predators, attacking prey that move within their sensory range. By reducing movement through the water column, *S. exspinosus* reduced its encounter rate with these predators (Havel et al. 1993). Body size for *S. exspinosus*, therefore, was probably less important in reducing predation than simply reducing its encounter rate. *Ceriodaphnia reticulata*, averaging a little over 0.5 mm, was probably below the electivity range of many larger late-season predators.

CONCLUSIONS

The cladoceran communities of temporary woodland ponds appear to follow a reasonably predictable succession of species. Seasonal changes in community structure and body sizes appear to be closely linked with shifts in the predator community. Although this study represents but one pond and one season, similar trends in succession and body size relationships were observed in less-intensive studies of 4 other woodland ponds in the Lansing, Michigan area. In two of these, *S. exspinosus* was replaced by *S. vetulus* (O.F.Müller) and *C. reticulata* was replaced by *C. quadrangula* (O.F.Müller). There appears to be a relatively small pool of potential cladoceran species capable of inhabiting these temporary woodland ponds.

This primarily descriptive study has raised some important questions regarding the cladoceran communities of temporary woodland ponds. Clearly, there is much research to be conducted on the zooplankton communities that occupy these relatively small, ephemeral habitats:

1. What are the cues that trigger ephippia production in *D. ephemeralis* and the larger form of *D. pulex*? Are they directly predator induced (unlikely for *D. ephemeralis* but certainly possible for *D. pulex*), or are they environmentally-induced?
2. What role do seasonal changes in the algal community play in cladoceran succession? Although it appears that predation plays the major role in shaping the cladoceran community, it is doubtful that food resources are not influential in some manner.
3. What is the genetic relationship between the early and late forms of *D. pulex*? If they are different genotypes, is the later form also present early in the season as well as the larger form? If it is not present early, what cue triggers its appearance?
4. What is the relationship of the larger filtering organisms such as mosquito larvae and fairy shrimp to the cladoceran community? Do they play an important role in nutrient cycling or are they simply competitors of cladocerans for algal resources?
5. What are the quantitative advantages of the two forms of *D. pulex* when exposed to their respective predators? Morphological traits infer certain advantages. Does the smaller, large-spined form have an advantage over the larger, small-spined form when exposed to salamander predation?

REFERENCES

Aly, C. 1988. Filtration rates of mosquito larvae in suspensions of latex microspheres and yeast cells. Entomologia Experimentalis et Applicata 46: 55-61.

Ameen, M. and T. M. Iversen. 1978. Food of *Aedes* larvae (Diptera: Culicidae) in a temporary forest pool. Archiv für Hydrobiologie 83: 552-564.

Arts, M. T., E. J. Maly, and M. Pasitschniak. 1981. The influence of *Acilius* (Dytiscidae) predation on *Daphnia* in a small pond. Limnology and Oceanography 26: 1172-1175.

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. Limnology and Oceanography 30: 1037-1045.

Black, A. R. and S. I. Dodson. 1990. Demographic costs of *Chaoborus*-induced phenotypic plasticity in *Daphnia pulex*. Oecologia 81: 117-122.

Brambilla, D. J. 1980. Seasonal changes in size at maturity in small pond *Daphnia*, p. 438-455. *In* [ed.], W.C.Kerfoot, Evolution and Ecology of Zooplankton Communities. New England University Press. Hanover, NH.

Brambilla, D. J. 1982. Seasonal variation of egg size and number in a *Daphnia pulex* population. Hydrobiologia 97: 233-248.

Brandl, Z. and C. H. Fernando. 1974. Feeding of the copepod *Acanthocyclops vernalis* on the cladoceran *Ceriodaphnia reticulata* under laboratory conditions. Canadian Journal of Zoology - Journal Canadien de Zoologie 52: 99-105.

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

Cox, E. J. 1996. Identification of Freshwater Diatoms From Live Material. Chapman and Hall. London.

Crosetti, D. and F. G. Margaritora. 1987. Distribution and life cycles of cladocerans in temporary pools from central Italy. Freshwater Biology 18: 165-175.

Dodson, S. I. 1974. Zooplankton competition and predation: An experimental test of the size-efficiency hypothesis. Ecology 55: 605-613.

Fryer, G. 1957. The food of some freshwater cyclopoid copepods and its ecological significance. Journal of Animal Ecology 26: 263-286.

Gliwicz, Z. M. 1990. Food thresholds and body size in cladocerans. Nature 343: 638-640.

Gliwicz, Z. M. and J. Pijanowska 1989. The role of predation in zooplankton succession, p. 253-296. *In* [ed.], U.Sommer, Plankton Ecology: Succession in Plankton Communities. Springer-Verlag. Berlin.

Gliwicz, Z. M. and H. Stibor. 1993. Egg predation by copepods in *Daphnia* brood cavities. Oecologia 95: 295-298.

Hairston, N. G., Jr. 1987. Diapause as a predator avoidance adaptation, p. 281-299. *In* [eds.], W.C.Kerfoot and A.Sih, Predation: Direct and Indirect Impacts on Aquatic Communities. University Press of New England. Hanover, N.H.

Hairston, N. G., Jr. and W. R. Munns, Jr. 1984. The timing of copepod diapause as an evolutionarily stable strategy. American Naturalist 123: 733-751.

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

Hall, D. J., S. T. Threlkeld, C. W. Burns, and P. H. Crowley. 1976. The size-efficiency hypothesis and the size structure of zooplankton communities. Annual Review of Ecology and Systematics 7: 177-208.

Hanazato, T. and S. I. Dodson. 1995. Morphological defenses of *Daphnia* against copepod predation on eggs. Archiv für Hydrobiologie 133: 49-59.

Harding, J. H. 1997. Amphibians and Reptiles of the Great Lakes Region. University of Michigan Press. Ann Arbor.

Havel, J. E., J. Link, and J. Niedzwiecki. 1993. Selective predation by *Lestes* (Odonata: Lestidae) on littoral microcrustacea. Freshwater Biology 29: 47-58.

Higgins, M. J. and R. W. Merritt 1999. Temporary woodland ponds in Michigan: Invertebrate seasonal patterns and trophic relationships, p. 279-297. *In* [eds.], D.P.Batzer, R.B.Rader, and S.A.Wissinger, Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley and Sons. New York.

Hobbie, J. E., R. J. Daley, and S. Jasper. 1977. Use of Nuclepore filters for counting bacteria by fluorescence microscopy. Applied and Environmental Microbiology 33: 1225-1228.

LaBerge, S. and B. J. Hann. 1990. Acute temperature and oxygen stress among genotypes of *Daphnia pulex* and *Simocephalus vetulus* (Cladocera, Daphniidae) in relation to environmental conditions. Canadian Journal of Zoology - Journal Canadien de Zoologie 68: 2257-2263.

Lehman, J. T. 1980. Nutrient recycling as an interface between algae and grazers in freshwater communities, p. 251-263. *In* [ed.], W.C.Kerfoot, Evolution and Ecology of Zooplankton Communities. University Press of New England. Hanover, NH.

Lockaby, B. G., A. L. Murphy, and G. L. Somers. 1996a. Hydroperiod influences on nutrient dynamics in decomposing litter of a floodplain forest. Soil Science Society of America Journal 60: 1267-1272.

Lockaby, B. G., R. S. Wheat, and R. G. Clawson. 1996b. Influence of hydroperiod on litter conversion to soil organic matter in a floodplain forest. Soil Science Society of America Journal 60: 1989-1993.

Luning, J. 1995. Life-history responses to *Chaoborus* of spined and unspined *Daphnia pulex*. Journal of Plankton Research 17: 71-84.

Lynch, M. 1978. Complex interactions between natural coexploiters--*Daphnia* and *Ceriodaphnia*. Ecology 59: 552-564.

Lynch, M. and J. Shapiro. 1981. Predation, enrichment, and phytoplankton community structure. Limnology and Oceanography 26: 86-102.

Modlin, R. F. 1982a. A comparison of two *Eubranchipus* species (Crustacea: Anostraca). American Midland Naturalist 107: 107-113.

Modlin, R. F. 1982b. Successional changes, variations in population densities, and reproductive strategies of Cladocera in two temporary ponds in north Alabama. Journal of Freshwater Ecology 1: 589-598.

Moore, M. V. 1988. Differential use of food resources by the instars of *Chaoborus punctipennis*. Freshwater Biology 19: 249-268.

Parejko, K. 1991. Predation by chaoborids on typical and spined *Daphnia pulex*. Freshwater Biology 25: 211-217.

Pennak, R. W. 1989. Fresh-Water Invertebrates of the United States: Protozoa to Mollusca, 3rd. ed. John Wiley and Sons. New York.

Pijanowska, J. 1990. Cyclomorphosis in *Daphnia*: An adaptation to avoid invertebrate predation. Intrazooplankton.Predation. 198 pp. 41-50: 41-50.

Porter, K. G. 1984. Natural bacteria as food resources for zooplankton, p. 341-345. *In* [eds.], M.J.Klug and C.A.Reddy, Current Perspectives in Microbial Ecology. American Society for Microbiology. Washington, D.C.

Porter, K. G. and Y. S. Feig. 1980. The use of DAPI for identifying and counting aquatic microflora. Limnology and Oceanography 25: 943-948.

Porter, K. G. and J. D. Orcutt, Jr. 1980. Nutritional adequacy, managability, and toxicity as factors thet determine the food quality of green and blue-green algae for *Daphnia*, p. 268-281. *In* [ed.], W.C.Kerfoot, Evolution and Ecology of Zooplankton Communities. University Press of New England. Hanover, NH.

Prescott, G. W. 1962. Algae of the Western Great Lakes, 2nd. ed. Wm. C. Brown. Dubuque, IA.

Prescott, G. W. 1978. How to Know the Freshwater Algae, 3rd. ed. Wm. C. Brown. Dubuque, IA.

Repka, S., M. Ketola, and M. Walls. 1994. Specificity of predator-induced neck spine and alteration in life history traits in *Daphnia pulex*. Hydrobiologia 294: 129-140.

Repka, S. and K. Pihlajamaa. 1996. Predator-induced phenotypic plasticity in *Daphnia pulex*: Uncoupling morphological defenses and life history shifts. Hydrobiologia 339: 67-71.

Repka, S., M. Walls, and M. Ketola. 1995b. Neck spine protects *Daphnia pulex* from predation by *Chaoborus*, but individuals with longer tail spine are at a greater risk. Journal of Plankton Research 17: 393-403.

Romanovsky, Y. E. and I. Y. Feniova. 1985. Competition among cladocera: Effects of different levels of food supply. Oikos 44: 243-252.

Schaffer, W. M. 1974. Optimal reproductive effort in fluctuating environments. American Naturalist 108: 783-790.

Schindler, J. E. 1970. Food quality and zooplankton nutrition. Journal of Animal Ecology 40: 589-595.

Schwartz, S. S. 1991. Predator-induced alterations in *Daphnia* morphology. Journal of Plankton Research 13: 1151-1161.

Schwartz, S. S. and P. D. N. Hebert. 1985. *Daphniopsis ephemeralis* sp.n. (Cladocera, Daphniidae): a new genus for North America. Canadian Journal of Zoology - Journal Canadien de Zoologie 63: 2689-2693.

Ślusarczyk, M. 1995. Predator-induced diapause in *Daphnia*. Ecology 76: 1008-1013.

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

Taylor, B. E., R. A. Estes, J. H. K. Pechmann, and R. D. Semlitsch. 1988. Trophic relations in a temporary pond: Larval salamanders and their microinvertebrate prey. Canadian Journal of Zoology - Journal Canadien de Zoologie 66: 2191-2198.

Threlkeld, S. T. 1979. Midsummer dynamics of 2 *Daphnia* species in Wintergreen Lake, Michigan. Ecology 60: 165-179.

Tilman, D., R. Kiesling, R. Sterner, S. S. Kilham, and F. A. Johnson. 1986. Green, bluegreen and diatom agae: Taxonomic differences in competitive ability for phosphorus, silicon and nitrogen. Archiv für Hydrobiologie 106: 473-485.

Wiggins, G. B., R. J. Mackay, and I. Smith. 1980. Evolutionary and ecological strategies of animals in annual temporary pools. Archiv für Hydrobiologie, Supplement 58: 97-206.

Williams, D. D. 1987. The Ecology of Temporary Waters. Croom Helm, Timber Press. Portland, OR.

Wilson, M. S. and H. C. Yeatman 1959. Free-living Copepoda, p. 735-861. *In* [eds.], W.T.Edmondson, H.B.Ward, and G.C.Whipple, Fresh-Water Biology. John Wiley and Sons. New York.

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

CHAPTER 4

PHENOLOGY, BODY SIZE, AND PREDATION IN A TEMPORARY WOODLAND POND

ABSTRACT

Temporary ponds are characterized by annual cycle of flooding and drying, which requires aquatic organisms that occupy these habitats to adapt to these conditions. In these ephemeral environments, there are no large overwintering predators, and the aquatic community is reset each spring. Organisms that are particularly well adapted to life in temporary woodland ponds include *Aedes* mosquito larvae, fairy shrimp, and the cladoceran *Daphnia ephemeralis*. All of these are filtering or gathering collectors that begin development as soon as the pond floods early in the spring. None of these organisms exhibit any strong morphological defenses to predation, despite a substantial predatory community that occupies these ponds during the course of the wet phase. As a consequence of beginning development early in the season, potential prey taxa such as these are exposed to fewer and smaller predators early than they would be if development was initiated later in the season. These organisms minimize predation early by being exposed to few predators, and later by reaching body sizes that are beyond the capturing/handling size of most available predators.

INTRODUCTION

Understanding the forces that shape community composition has been of interest to ecologists for many years. Aside from the physical and nutritional requirements necessary for organisms to survive, biotic interactions among and within species have been viewed as a major driving force in community organization. While competition has traditionally been viewed as being of primary importance to community structure (e.g., Tilman 1982), more recently, the importance of predation has been emphasized, particularly in aquatic environments (Connell 1975; Zaret 1980; Kerfoot and Sih 1987; Sih 1987; Wellborn et al. 1996; Schneider 1997). The term "predation" is used here in the traditional sense meaning one animal consuming another, and does not include aquatic herbivory (Brönmark and Hansson 1998).

Zaret (1980) recognized two types of predators in aquatic communities, gape-limited and size-dependent predators. Gape-limited predators essentially capture prey with their mouth and are therefore limited in the size of prey they can capture by the size of their gape. Size-dependent predators do not ingest prey whole, and are limited to the size of prey that they can effectively capture and handle. Planktivorous fish and salamander larvae (e.g., *Ambystoma*) are examples of gape-limited predators reported by Zaret (1980). I would also include dytiscid beetle larvae in this category as they capture prey with their mandibles and subsequently suck fluids from their prey. Examples of size-dependent predators given by Zaret (1980) include phantom midge larvae (*Mochlonyx* and *Chaoborous*), which capture prey with modified antennae. It is important to note that in both of these categories of predators, potential prey that exceed the size limitations are effectively immune to capture. Large prey body size relative to predators can be a significant defense to lessen or even eliminate predation risk, particularly in invertebrate predator/prey relationships. Cohen et al. (1993) examined body sizes of predators and prey in 70 different communities and found a strong positive correlation for size in invertebrate predator/prey relationships, with most predators consuming prey that were equal to or smaller than them in size (Peters 1983; Cohen et al. 1993).

The concept of enemy free space (Jeffries and Lawton 1984) has been used to describe the role of predation in shaping ecological niches, in both contemporary and evolutionary time frames. Enemy free space refers to any trait that reduces or eliminates a species' vulnerability to predation (Jeffries and Lawton 1984). Such traits may be behavioral, morphological, physiological, and even phenological. Predation involves three components: detection, pursuit, and capture (Holling 1966), and a given prey's defense may be involved in any of these. Defensive traits are not mutually exclusive, and a given species may exhibit more than one, particularly in different life stages. Connell (1975) recognized the potential advantage for a prey species to outgrow its predators, but also noted that such a defense is less effective in younger (smaller) individuals. By employing other defensive traits in the more vulnerable early life stages, however, outgrowing potential predators may be an effective defense.

Temporary ponds are ideal habitats in which to study seasonal changes in predator-prey relationships. Due to the dry phase of these ponds, the aquatic communities within them are "reset" each year. In temporary woodland ponds in Michigan, there are no large overwintering predators. Flooding in the spring triggers a succession of species, with most of the earliest species hatching from resting eggs. Many predators appear later in the spring, either hatching from resting eggs or arriving as immigrants from permanent water refuges (Wiggins et al. 1980; Higgins and Merritt 1999). Among the earliest to appear in the spring are *Aedes* mosquito larvae, *Eubranchipus* fairy shrimp, and the cladoceran, *Daphnia ephemeralis*. All of these organisms are extremely well adapted to life in temporary ponds. They all have freeze and drought-resistant eggs and are capable of surviving and growing in the cold water temperatures ice cover of early spring (Wiggins et al. 1980; Westwood et al. 1983; Schwartz and Hebert 1985). None of these organisms, however, has any particular morphological adaptation against predators. Because predators are very abundant in temporary pond communities (Higgins and Merritt 1999), the general lack of morphological defenses to reduce predation suggests that other means of reducing or eliminating predation risk are more important.

I hypothesize that organisms that are well adapted to temporary habitats reduce their exposure to predators by beginning development early and by reaching body sizes that are larger than can be effectively handled by most available predators. Such a strategy can be effective only in environments like temporary ponds, where the communities must restart each season and where there are no large overwintering predators.

MATERIALS AND METHODS

Study Site

Research was conducted at a small woodland pool, Wild Ginger #1 (WG-1), near Haslett, Michigan, USA (42° 45' 30"N, 84° 23' 50"W). This pond is approximately 600 m^2 when completely flooded, with a maximum depth of 0.8 m. In an average year, the pond begins to fill with water during the late autumn, and reaches its maximum size and depth in March from snowmelt and rainfall. The pond loses all surface water between mid July and early August. Water temperatures range from < 5° C in March to > 25° C in June and July. The basin is situated within a 60-70 year-old deciduous woodlot of 70 ha and is surrounded by trees and shrubs, primarily red maple (*Acer rubrum*), American elm (*Ulmus americana*), black ash (*Fraxinus nigra*), swamp white oak (*Quercus bicolor*), and northern swamp dogwood (*Cornus racemosa*). The canopy does not completely close over the pond, but after the trees leaf out, most sunlight reaching the surface is indirect. Leaves from trees near the basin and the surrounding upland (predominantly sugar maple and white oak) contribute to the extensive detritus that provides the energy source for the temporary pond community in the spring (Higgins and Merritt 1999).

Field Studies

In order to examine successional trends, the aquatic invertebrate community was sampled every 2 weeks in 1996-1998, from the time ice began to melt along the margins of the pond until it dried, usually March-July. Samples were taken with a D-frame aquatic net (mesh size 0.5 mm) and a standard 500 ml mosquito dipper. Samples were preserved in 70% ethyl alcohol and identified using keys in Merritt and Cummins (1996) and Pennak (1989). Individual body lengths of both predators and prey were measured with an ocular micrometer on a dissecting microscope to the nearest 0.1 mm. Because predator and prey size was being considered, each larval instar of the insects was treated as a separate predator or prey organism in this study. Thus, if two larval instars of the same predatory species were present in the pond at the same time, there would most likely be differences in electivity of prey size between them and were therefore treated as different predators.

Zooplankton samples were collected weekly in 1999. A plankton net (mesh size 80 µm) was towed 3 times on each sampling date just below surface of the pond for a distance of 1.5 m. In the early spring, the net could not be towed due to ice cover, nor could it be towed in shallow water near shore. To sample near shore and during ice cover, a standard 500 ml mosquito dipper was used to collect water, which was then poured through the plankton net. This procedure was repeated 4 times for a total volume of 2 l at each of 3 sampling locations on each date. All samples were preserved in 70% ethanol and specimens were identified using keys in Pennak (1989) and Wilson and Yeatman (1959). Cladoceran body lengths were measured to the nearest 0.03 mm using an ocular micrometer on a dissecting microscope. Body lengths for cladocerans do not include tail spines.

Predator/Size Experiments

To determine prey size limitations of various predators, laboratory experiments were conducted using *Aedes stimulans* (Walker) larvae as prey, with the predators *Mochlonyx*, *Acilius*, and *Agabus* larvae. The purpose of these experiments was not to duplicate natural conditions of predator versus prey, but to determine the upper limit of prey size for different stages of various predators. In order to simultaneously obtain different larval instars of mosquitoes, first instars were collected in the field and reared at different temperatures to accelerate or retard their growth. Only 24 third-instar *Agabus* were collected, so only 8 replicates were used for that particular experiment. All other experiments used 10 replicates. Predators were field collected as needed and starved for 6 hours before each experiment. In experiments with the larval phantom midge, *Mochlonyx*, 4 *Aedes* larvae were placed in each of 10, 30-ml vials with 20 ml of filtered (125 µm) pond water along with 0.5 ml solution of finely-ground TetraMin® fish food (0.1g/25 ml water) as food for the mosquitoes. Each vial then received one *Mochlonyx* larvae in the following combinations: first-instar *Mochlonyx* separately with first and second-instar *Aedes*, second-instar *Mochlonyx* separately with first and second-instar *Aedes*, and third-instar *Mochlonyx* separately with second and third-instar *Aedes*.

Experiments using the dytiscid beetle larvae, *Acilius* and *Agabus* were conducted in a similar manner using 60 ml plastic cups with 40 ml filtered pond water. Each cup received 4 mosquito larvae and one predator in the following combinations: first-instar *Acilius* separately with second and fourth-instar *Aedes*, second-instar *Acilius* separately with second and fourth-instar *Aedes*, first-instar *Agabus* separately with first, second, and third-instar *Aedes*, second-instar *Agabus* separately with first, second, and third-instar *Aedes*, and third-instar *Agabus* separately with second, third, and fourth-instar *Aedes*.

All experiments were conducted at a constant temperature of 15° C in an environmental chamber with 12:12 h light:dark regime. After 48 h the number of *Aedes* larvae eaten within each container was recorded.

Crop contents of fourth-instar *Mochlonyx* larvae were also examined. Larvae were captured in the field with a mosquito dipper, immediate killed by placing them in hot water, and then transferred to 70% ethyl alcohol. The crop was removed intact from each larva with minuten pins and transferred to a clean microslide. A drop of water was added and the crop contents were teased apart and viewed at 100x and 250x.

RESULTS

Predator Succession

Very early in the season few predators were present (Figure 1). When ice on the pond first began to melt, the only predators collected were the cyclopoid copepods, *Diacyclops bicuspidatus thomasi* (Forbes) and *Acanthocyclops vernalis* (Fischer).

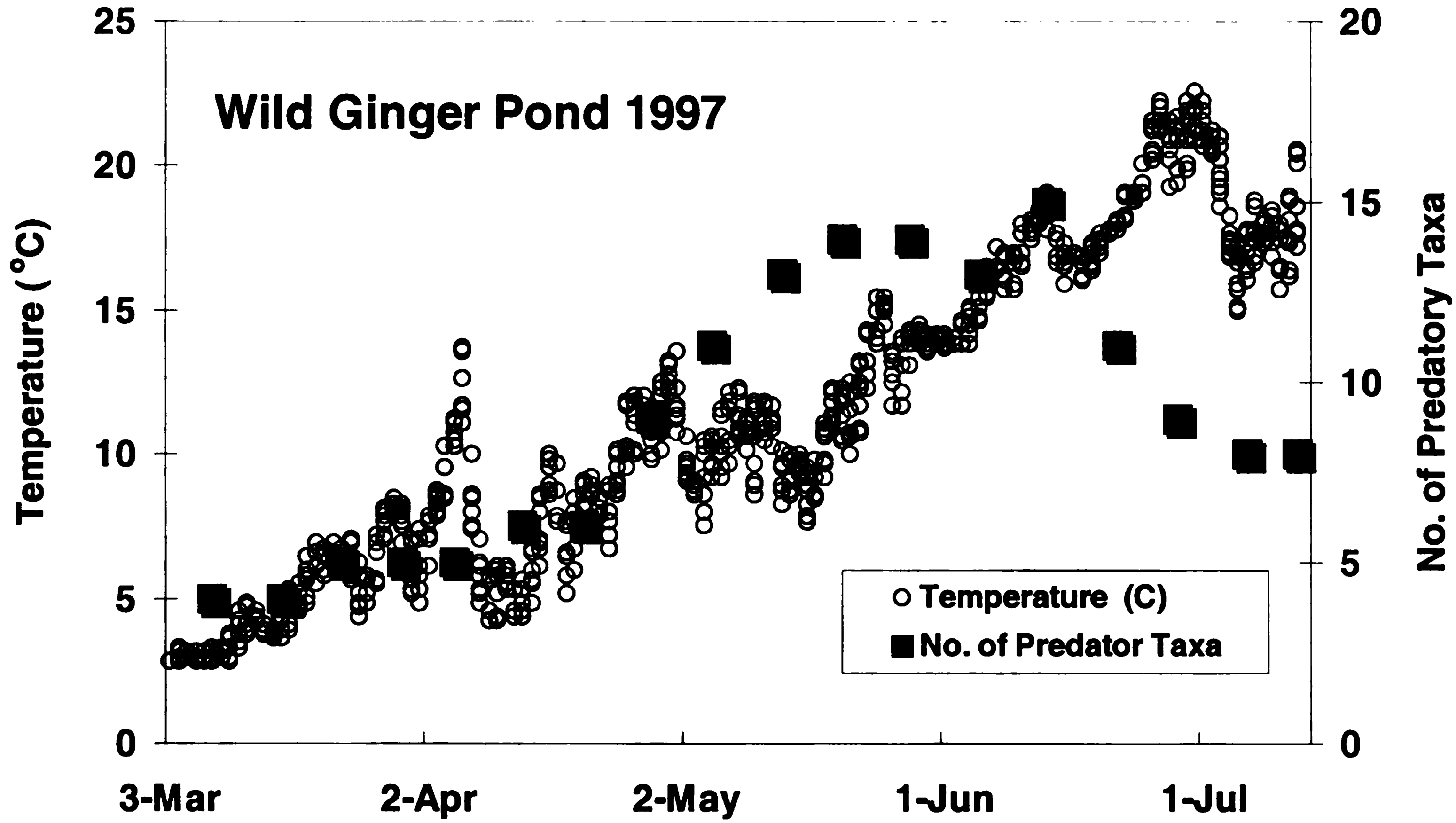

Figure 1. Relationship of seasonal changes in number of predatory taxa with changes in water temperatures, Wild Ginger #1 Pond.

Shortly thereafter, these were joined by the larger cyclopoid, *Macrocyclops albidus* (Jurine), and first instars of the phantom midge, *Mochlonyx cinctipes* (Coquillett). The 3 cyclopoid copepod species were not collected beyond the end of April, but another, smaller cyclopoid, *Diacyclops navus* (Herrick) was present in May and June. *Mochlonyx* larvae (third and fourth instars) were present throughout April, emerging as adults in early May. As *Aedes* mosquito larvae began hatching in substantial numbers when the pond reached maximum size with early spring rain, first instars of the dytiscid beetle, *Agabus erichsoni* (Gemminger and Harold) also appeared in collections. This species was most often found clinging to detritus near the pond's margins. This beetle developed rapidly, completing larval development by mid to late April.

Odonate larvae of *Lestes* (primarily *dryas* Kirby) and *Sympetrum* (mostly *obtrusum* Hagen) began appearing in collections in April. Teneral *Lestes* and *Sympetrum* adults were observed along the margins of the pond by mid to late June. Phantom midge larvae, *Chaoborus americanus* (Johannsen) were collected from mid-late April, and *Chaoborus* larvae, representing multiple generations, were present in the pond for the remainder of the season. A surprisingly significant predator of cladocerans in temporary woodland ponds is the dytiscid beetle larva, *Acilius semisulcatus* Aubé, which was first recovered in late April through early May, and was abundant through the entire month of May. Unlike many other dytiscid larvae that are found clinging to debris near the margins of ponds, first and second instars of *A. semisulcatus* were most often found near the surface in the deeper areas of the ponds. The larvae suspend themselves within the water column with their respiratory spiracles in contact with the surface and capture prey with their mandibles. *Acilius* larvae were found to be important predators of *Daphnia pulex* in a small pond near Montreal (Arts et al. 1981).

Larvae of salamanders (*Ambystoma* spp.) were present in WG-1 every spring in 1996-1999, although their abundance varied considerably from year to year. Salamander larvae were abundant in 1996 and 1997, but occurred in very low numbers in 1998 and 1999. Adult salamanders enter small woodland pools in late March or early April to breed and lay eggs, and the eggs hatch in 3-5 weeks, depending on temperature (Harding 1997). Larvae of *Ambystoma* are the only important vertebrate predators in small, temporary woodland ponds like WG-1, and they can have a significant impact on a zooplankton community as size-selective predators (Taylor et al. 1988).

Several predatory larvae appeared later in the spring, most notably hydrophilid beetles (*Hydrochara*), dytiscid beetles (*Dytiscus* and *Laccophilus*), notonectids (*Notonecta*), veliids (*Microvelia*), and in some years belostomatids (*Belostoma*). All of these predators are migrant species, the adults of which overwinter in permanent water and recolonize temporary ponds in the spring to breed (Wiggins et al. 1980).

Predation and Body Size

The results of the predator/size experiments are summarized in Table 1. For *Mochlonyx*, it is clear that first and second larval instars are capable of catching and consuming first and second instar *Aedes* larvae that are less than or equal to them in length. After the second instar, *Aedes* larvae were apparently too large for *Mochlonyx* larvae to capture. Although fourth-instar *Mochlonyx* were not used in any of these experiments, examination of gut contents from 20 field-collected fourth-instar *Mochlonyx* failed to identify any mosquito larvae. First-instar *Acilius* larvae were

Table 1. Predator feeding experiments using 4 larval *Aedes stimulans* mosquitoes and 1 larval predator. Values are percentages of mosquitoes eaten and mean number consumed per container in parentheses; n= 10 for all except third instar *Agabus* (n=8). Mean length of each instar is given in parentheses.

Predator **Instar #**	***Aedes stimulans*** **Instar #** **1st** (1.9)	**2nd** (4.2)	**3rd** (7.3)	**4th** (9.7)
Mochlonyx				
1st (2.1)	15.0 (0.6)	0	---	---
2nd (3.8)	97.5 (3.9)	22.5 (0.9)	---	---
3rd (5.2)	---	72.5 (2.9)	0	---
Acilius				
1st (8.4)	---	95.0 (3.8)	---	0
2nd (14.8)	---	67.5 (2.7)	---	50.0 (2.0)
Agabus				
1st (7.4)	100 (4.0)	95.0 (3.8)	20.0 (0.8)	---
2nd (11.7)	80.0 (3.2)	100 (4.0)	77.5 (3.1)	---
3rd (18.2)	---	82.5 (3.3)	81.25 (3.25)	72.5 (2.9)

capable of capturing up to second-instar *Aedes* larvae, but fourth-instar mosquitoes were apparently beyond their handling size. Second-instar *Acilius* larvae, however, were quite capable of capturing fourth-instar *Aedes*. The larvae of *Agabus erichsoni* were capable of capturing a wide range of sizes of mosquito larvae in all three instars. Even first-instar *Agabus* could at least occasionally capture third-instar *Aedes* larvae (Table 1).

Seasonal Changes in Body Size Relationships

Early in the season, very few predators were present in the pond. At the time that *Aedes* larvae and *Eubranchipus* hatched, the predators were first instar *Mochlonyx* and *Agabus* beetle larvae, and cyclopoid copepods, the largest being *Macrocyclops*. With the exception of *Agabus*, all these predators were smaller than or equal to the two prey taxa in size (Figure 2). As the prey grew, they reached sizes larger than all but *Agabus*. Predatory taxa that appeared later in the spring were all smaller than *Aedes* and *Eubranchipus*, with the exception of first instar *Dytiscus* larvae, which was first collected in late April. By this time, *Eubranchipus* had completed its life cycle. *Dytiscus* larvae were never abundant in WG-1 but they are among the largest beetle larvae (first instars are 14 mm long) and therefore may be relatively significant predators (Young 1967). These beetles had abundant fourth instar *Aedes* larvae on which they could feed until the mosquitoes pupated and emerged in early May. After that, it appeared that *Dytiscus* larvae preyed largely upon tadpoles of wood frogs. By the time more predators arrived, and others reached larger size, the mosquitoes had emerged as adults.

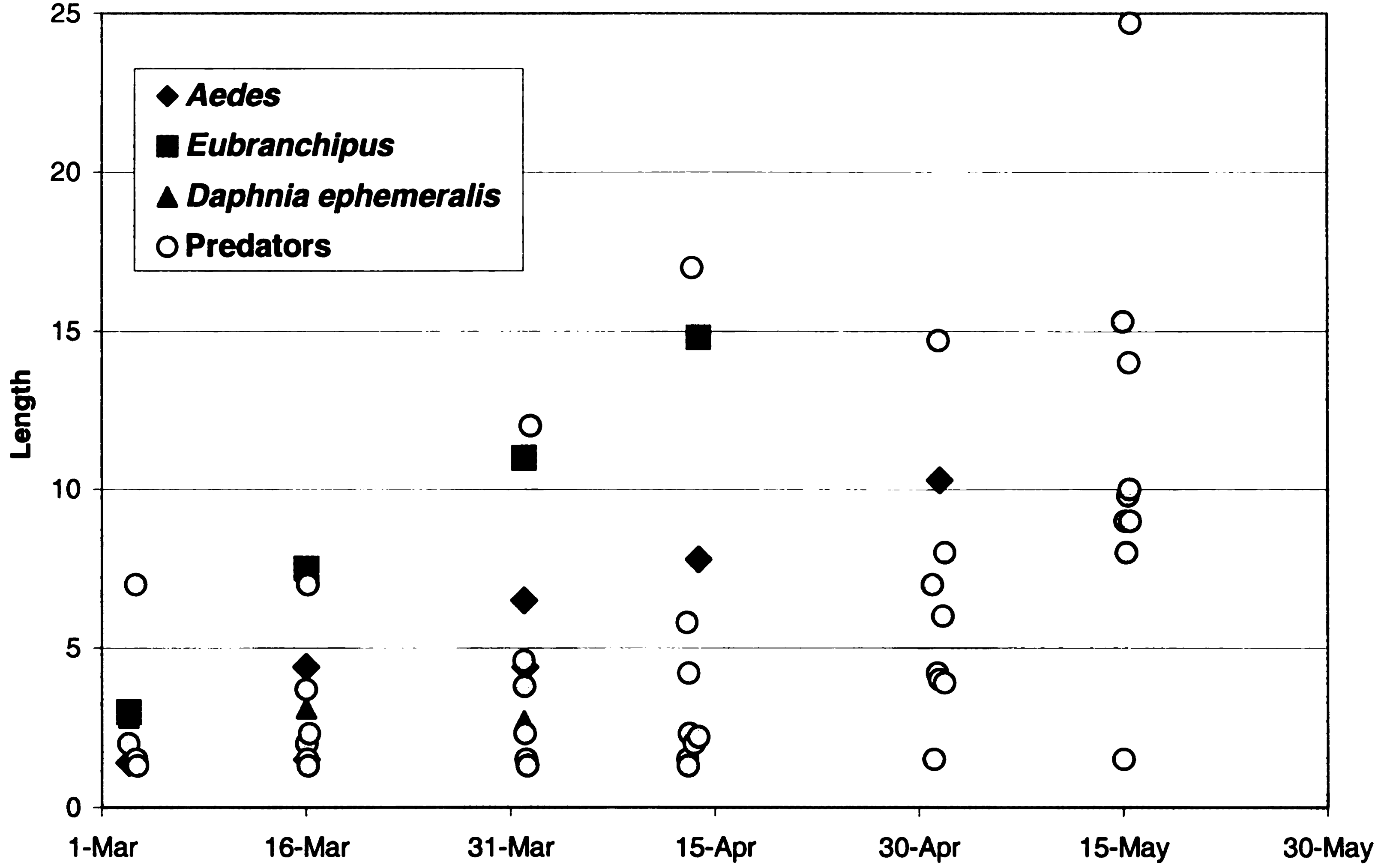

Figure 2. Seasonal trends in body size relationships between predators and three prey taxa, Wild Ginger #1 Pond.

The situation was very similar with the cladoceran community in WG-1 (Figure 3). Very early in the spring, the only species present was *Daphnia ephemeralis*, and parthenogenetic females reached lengths of 3 mm. Predatory copepods and early larval instars of *Mochlonyx* would necessarily prey on much smaller individuals during this time, with adult *D. ephemeralis* effectively immune to predation due to their large size. Juvenile *D. ephemeralis* exhibit a distinct tail and neck spine, while spines are absent in adults (Schwartz and Hebert 1985). Tail and neck spines in cladocerans are anti-predator morphological traits that are often inducible in the presence of predators (Luning-Krizan 1997; Kolar and Wahl 1998). *Daphnia ephemeralis* began sexual reproduction and the production of ephippia in early April, and completed its life cycle by the end of April, before the appearance of most of the larger predators.

Following *D. ephemeralis* in succession, *Daphnia pulex* began to appear in collections in early April. Sharing the same predators as *D. ephemeralis*, adult females achieved lengths up to 2.5-3.0 mm during April. In early May, however, males and sexually-reproducing females dominated the population, with the subsequent production of ephippia completed by the end of May. Parthenogenetic females collected during May were smaller--rarely exceeding 2.0 mm in length--and exhibited longer tail spines than adults collected in April (Figure 4). Only the smaller form of *D. pulex* was present through the remainder of the season. The initiation of sexual reproduction and the production of ephippia in *D. pulex* coincided with the appearance of two important predators in WG-1. Larvae of the dytiscid beetle, *Acilius semisulcatus* and salamanders, *Ambystoma* spp., both hatched in early May. Unlike the early spring predators, both of these organisms show a preference for larger prey (Brambilla 1980;

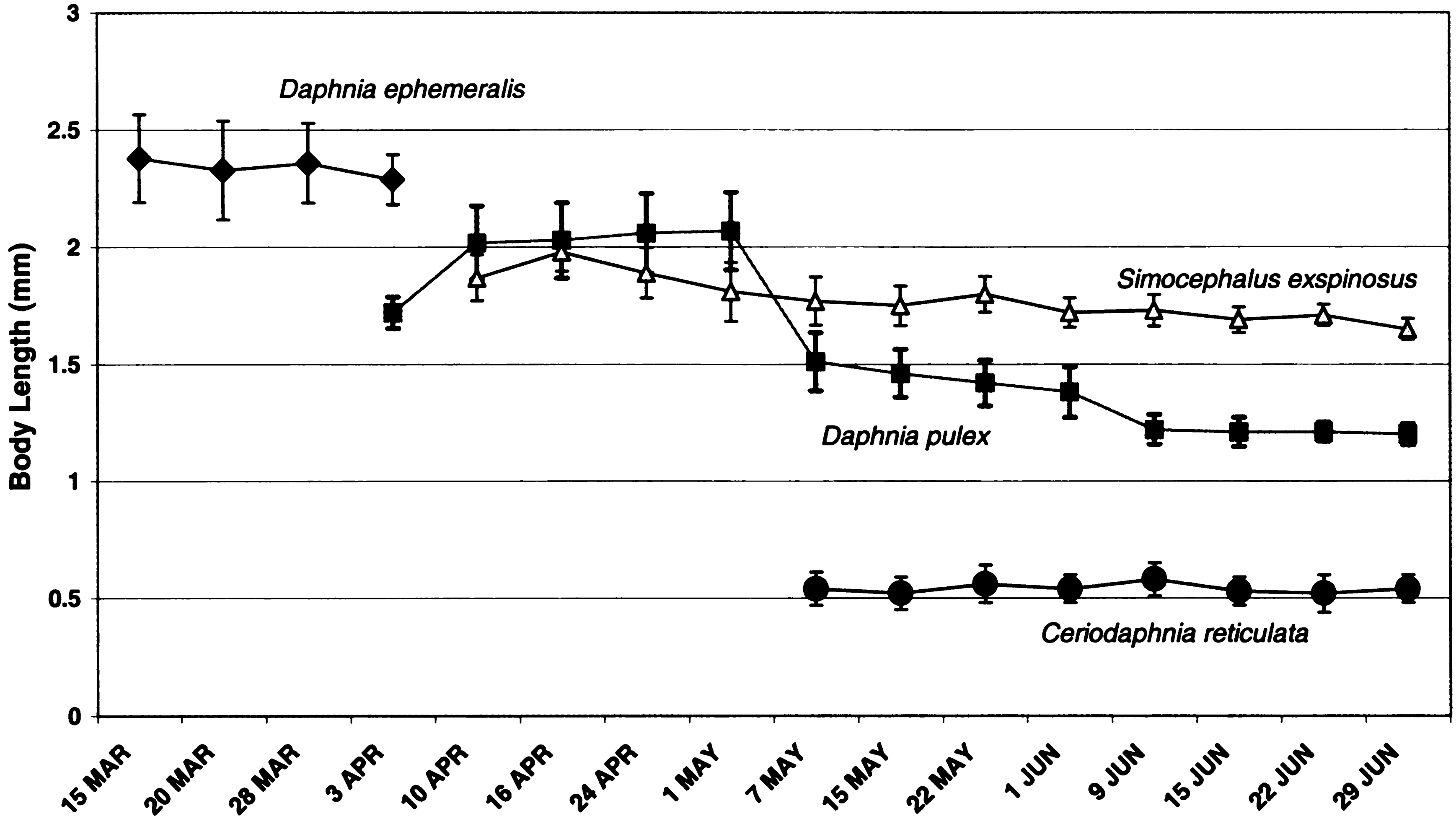

Figure 3. Seasonal changes in body lengths of parthenogenetic females of the four most abundant cladoceran species. Values are means (S.E.M.).

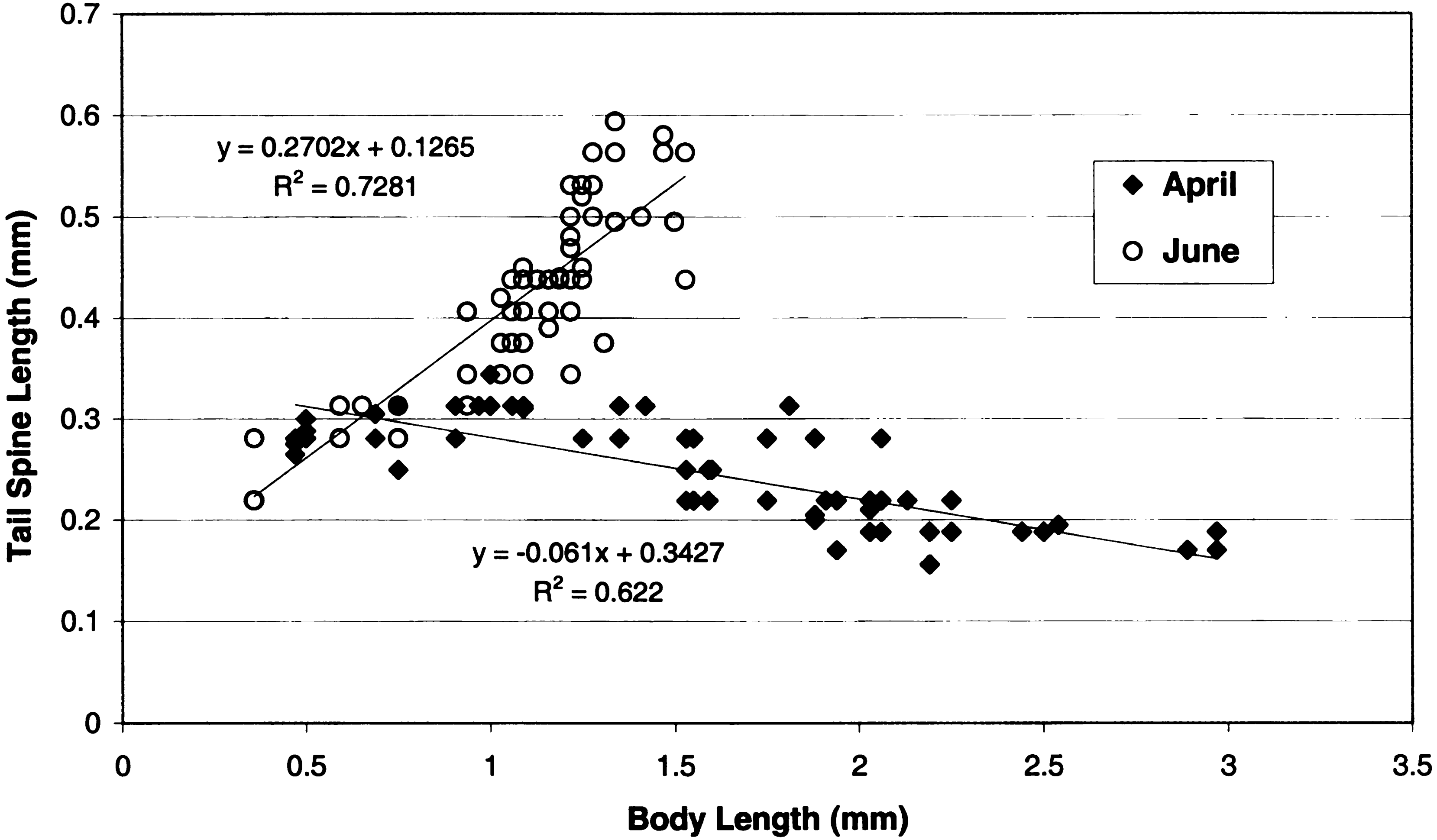

Figure 4. Tail spine length as a funtion of body length for parthenogenetic Daphnia pulex during early (April) and late (June) season. Slopes differ significantly at $p < 0.0001$.

Arts et al. 1981). Following their appearance in the pond, adult parthenogenetic females were much smaller, a size reduction that does not coincide with any reduction in food supply (see Chapter 3). In addition, tail lengths for the smaller form were longer, and increased with body length, compared to individuals collected in April, which showed a negative relationship between body size and tail length (Figure 4). This would suggest heavier predation pressure on larger individuals later as compared to earlier in the season.

DISCUSSION

In these temporary ponds, there is a definite trend for smaller and fewer predators early in the season than later in the season. Also, based on the laboratory feeding experiments, there are size limitations for most potential predators of *Aedes*. When *Aedes* and *Eubranchipus* hatch early in the spring, the only important predators are *Agabus* beetle larvae, *Macrocyclops* copepods, and perhaps early-instar *Mochlonyx* larvae. Although *Mochlonyx* has been implicated as an important predator of mosquito larvae (Morrison and Andreadis 1992), laboratory experiments indicate that mosquitoes can quickly outgrow these predators. These results are similar to those reported by O'Connor (1959) who found no mosquito remains in the crop contents of 30 third- and fourth-instar *Mochlonyx*. Although no feeding experiments were conducted with *Macrocyclops*, these predators would likely be important only during the first instar of the mosquitoes. In addtion, *Macrocyclops* density was very low ($0.1\ l^{-1}$) and they had other prey available, such as *Mochlonyx*, *Daphnia ephemeralis*, and other copepods. The copepod predators, in turn, could also fall prey to later-instar *Mochlonyx*. Only *Agabus erichsoni* larval development closely followed that of *Aedes*. This species is the earliest dytiscid beetle larvae to appear in temporary woodland ponds (James 1961), overwintering in the egg stage (James 1969; Wiggins et al. 1980). It is apparently an important predator of mosquito larvae in the spring. A laboratory-reared *A. erichsoni* consumed a total of 252 *Aedes* larvae during the course of its larval development (James 1969).

The cladoceran community also seems to be heavily influenced by predation. Early in the season, *Daphnia ephemeralis* is the only species present. Adults are large with little morphological defenses to predation (they even lack a tail spine), while juveniles and smaller sexual forms exhibit both neck and tail spines. *Daphnia ephemeralis* completes its life cycle early in the spring, prior to the arrival of larger predators. *Daphnia pulex*, which followed *D. ephemeralis* in succession, initially appeared as large adults with minimal tail spines. The appearance of larger predators such as *Ambystoma* salamanders and *Acilius* beetle larvae may have triggered the intense period of ephippia production by *D. pulex* in May. Predator-induced diapause is not unprecedented (Hairston and Munns 1984; Hairston 1987; • lusarczyk 1995). Thereafter, parthenogenetic *D. pulex* were smaller and possessed long tail spines.

Schneider and coworkers (Schneider and Frost 1996; Schneider 1997; Schneider 1999) have examined the role of pond duration in shaping temporary pond communities. Long-duration ponds support a much more diverse and, on average, longer-lived predator community than do short-duration ponds. Such predator communities may exclude certain taxa from long-duration ponds, or limit population sizes of other taxa (Schneider and Frost 1996; Schneider 1997). The mechanisms for such exclusion or limitation may include behavioral tradeoffs (Wellborn et al. 1996).

Animals in temporary habitats need to grow quickly and thus forage actively. This foraging activity, however, results in increased exposure to predation. In short-duration ponds with limited predator communities, this may not present a problem. In long-duration and permanent ponds, however, such active foraging may lead to extermination by predators. Thus, animals that are well adapted to temporary ponds are often excluded from permanent and semi-permanent ponds (Wellborn et al. 1996). Such mechanisms are viewed as accessory to life history constraints such as a prerequisite cold and dry period to initiate egg hatching (e.g., most *Aedes* mosquitoes). That many permanent and semi-permanent pond communities contain many temporary pond taxa and a reduced predator community after a drought (Jeffries 1994; Schneider and Frost 1996) supports the argument that such taxa are excluded by predation.

Animals such as *Aedes* and *Eubranchipus* are common in both short- and medium-duration temporary ponds. In small, short-duration ponds, there is little or no predation pressure. In medium-duration ponds like WG-1, predators are relatively scarce early, but increase in both numbers and size later in the spring. That mosquito larvae and fairy shrimp begin development very early in the spring in both types of ponds suggests that this life history trait probably evolved as a consequence of the ephemeral nature of the habitat and the need to develop rapidly in a shrinking environment. In medium-duration ponds, early development and relatively rapid growth have also served to minimize predation potential. *Aedes*, *Eubranchipus*, and *Daphnia ephemeralis* all begin development before most predators have appeared, and quickly outgrow the effective handling size of most available predators. By the time predators become abundant, *Eubranchipus* and *D. ephemeralis* have entered a diapausing stage and *Aedes* have exited the pond as adults. Only one major predator, the dytiscid beetle *Agabus erichsoni*, seems to have successfully circumvented this "exaptation" (Gould and Vrba 1982) by adapting--as its prey has--to the severe physical constraints of pond drying and the cold temperatures of early spring.

In addition to minimizing predation in time and through larger body size, there is another advantage to beginning development early that has not been previously discussed. Given the same sized predator, it is more advantageous for a prey population to be exposed to this predator later in its development rather than earlier, assuming that both stages are within the handling capabilities of the predator. A predator feeding on small prey will consume more individuals in order to become satiated than it will when feeding on large prey, i.e., predation rate falls with increasing size of prey (Travis et al. 1985). Thus, the same predator will have less of an impact on a prey species that is in the later stages of development rather than earlier stages because it will consume fewer large individuals. Potential prey organisms, such as *Aedes* mosquito larvae in temporary ponds, begin development early, and by doing so reduce their exposure to later-appearing predators. If late-instar larvae are exposed to predators (e.g., first-instar *Dytiscus* or second-instar *Acilius* beetle larvae), the impact on the population will be less than if these predators were preying on earlier instars. Lest I be accused of invoking group selection, predation rates for the prey population would decline with increasing body size, as would an *individual's* chance of being eaten.

A Model for Predator-Prey Relationships in Temporary Ponds

Predator-prey trends and relationships in temporary woodland ponds are summarized graphically in Figure 5. All of the biotic interactions are constrained by the physical

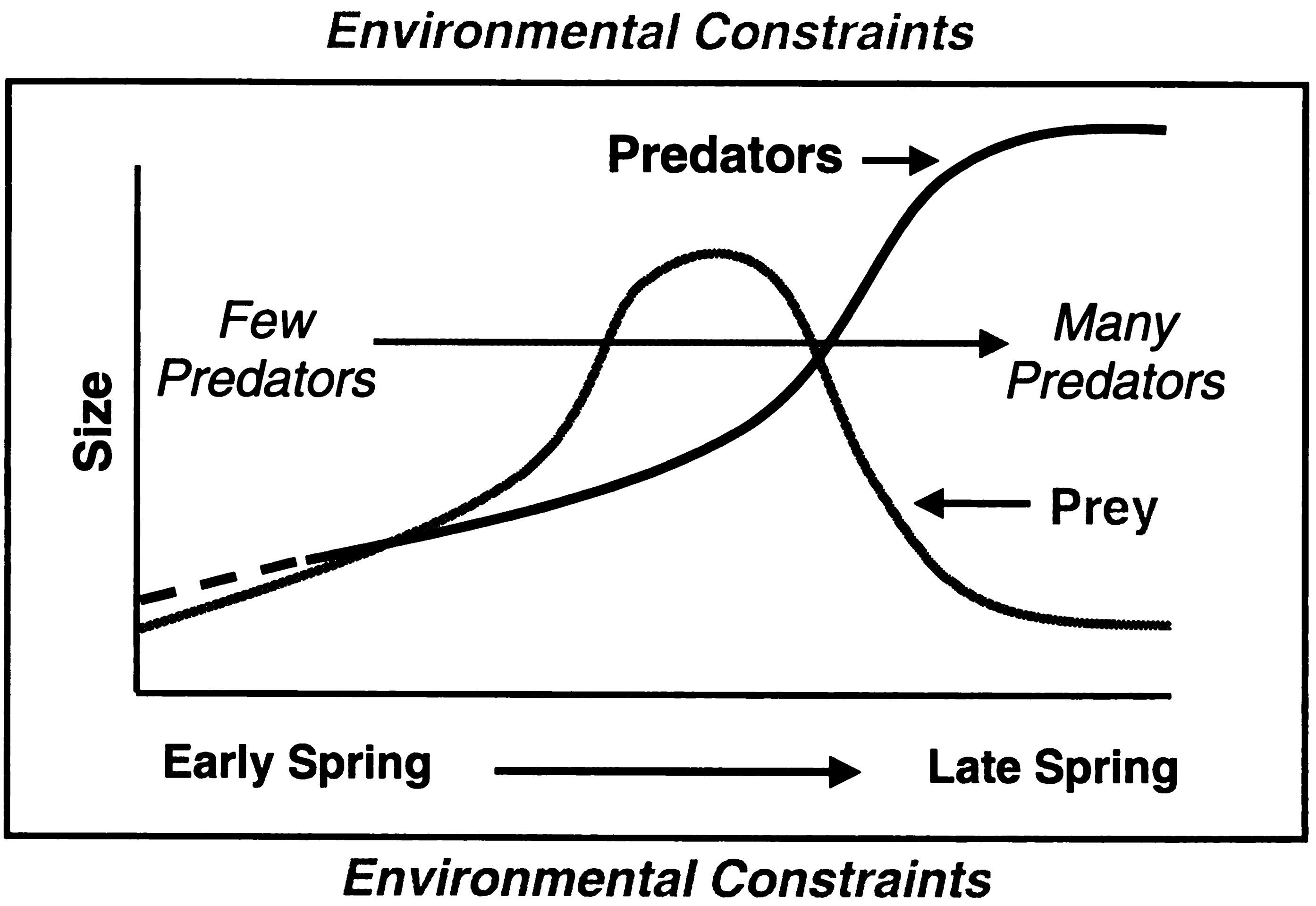

Figure 5. Generalized relationships of predator/prey phenology and body size in temporary woodland ponds.

environment (e.g., pond drying and cold temperatures early in the season). Species that are well adapted to temporary ponds have overcome the difficulties imposed by these environmental constraints and hatch very early in the season. The few predators that are present at that time are mostly small, and potential prey species can reduce predation risk by reaching a body size that is beyond the handling capabilities of most predators. Later in the season, predators are generally larger in size, and taxa like mosquito larvae, fairy shrimp, and *Daphnia ephemeralis* enter diapause, or exit the pond as adults in the case of mosquitoes, transforming to life history stages that are immune to aquatic predation. Both environmental constraints and predation, however, also can act as negative feedback to prey body size. Although large body size can reduce larval predation and could ultimately lead to increased adult fecundity, it can also increase the duration of the larval stage (Peters 1983). In an ephemeral habitat, such a tradeoff could potentially lead to increased larval mortality by not reaching maturity before the pond dried. Also, an increase in the duration of the larval stage may expose later stages of a prey species to larger predators that appear later in the season, thus negating any benefit of body size.

REFERENCES

Arts, M. T., E. J. Maly, and M. Pasitschniak. 1981. The influence of *Acilius* (Dytiscidae) predation on *Daphnia* in a small pond. Limnology and Oceanography 26: 1172-1175.

Brambilla, D. J. 1980. Seasonal changes in size at maturity in small pond *Daphnia*, p. 438-455. *In* [ed.], W.C.Kerfoot, Evolution and Ecology of Zooplankton Communities. New England University Press. Hanover, NH.

Brönmark, C. and L.-A. Hansson. 1998. The Biology of Lakes and Ponds. Oxford University Press. New York.

Cohen, J. E., S. L. Pimm, P. Yodzis, and J. Saldaña. 1993. Body sizes of animal predators and animal prey in food webs. Journal of Animal Ecology 62: 67-78.

Connell, J. H. 1975. Some mechanisms producing structure in natural communities: a model and evidence from field experiments, p. 460-490. *In* [eds.], M.L.Cody and J.M.Diamond, Ecology and Evolution of Communities. Belknap Press. Cambridge, Mass.

Gould, S. J. and E. S. Vrba. 1982. Exaptation--a missing term in the science of form. Paleobiology 8: 4-15.

Hairston, N. G., Jr. 1987. Diapause as a predator avoidance adaptation, p. 281-299. *In* [eds.], W.C.Kerfoot and A.Sih, Predation: Direct and Indirect Impacts on Aquatic Communities. University Press of New England. Hanover, N.H.

Hairston, N. G., Jr. and W. R. Munns, Jr. 1984. The timing of copepod diapause as an evolutionarily stable strategy. American Naturalist 123: 733-751.

Harding, J. H. 1997. Amphibians and Reptiles of the Great Lakes Region. University of Michigan Press. Ann Arbor.

Higgins, M. J. and R. W. Merritt 1999. Temporary woodland ponds in Michigan: Invertebrate seasonal patterns and trophic relationships, p. 279-297. *In* [eds.], D.P.Batzer, R.B.Rader, and S.A.Wissinger, Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley and Sons. New York.

Holling, C. S. 1966. the functional response of invertebrate predators to prey density. Memoirs of the Entomological Society of Canada 47: 3-86.

James, H. G. 1961. Some predators of *Aedes stimulans* (Walk.) and *Aedes tricurus* (Dyar) (Diptera: Culicidae) in woodland pools. Canadian Journal of Zoology - Journal Canadien de Zoologie 39: 533-540.

James, H. G. 1969. Immature stages of five diving beetles (Coleoptera: Dytiscidae), notes on their habits and life history, and a key to aquatic beetles of vernal woodland pools in southern Ontario. Proceedings of the Entomological Society of Ontario 100: 52-97.

Jeffries, M. 1994. Invertebrate communities and turnover in wetland ponds affected by drought. Freshwater Biology 32: 603-612.

Jeffries, M. J. and J. H. Lawton. 1984. Enemy free space and the structure of ecological communities. Biological Journal of the Linnean Society 23: 269-286.

Kerfoot, W. C. and A. Sih.Kerfoot, W. C. and Sih, A. [eds.] 1987. Predation: Direct and Indirect Impacts on Aquatic Communities. University Press of New England. Hanover, N.H.

Kolar, C. S. and D. H. Wahl. 1998. Daphnid morphology deters fish predators. Oecologia 116: 556-564.

Luning-Krizan, J. 1997. Neck-teeth induction in *Daphnia hyalina* under natural and laboratory conditions. Archiv für Hydrobiologie 140: 367-372.

Merritt, R. W. and K. W. Cummins (eds.). 1996. An Introduction to the Aquatic Insects of North America, 3rd. ed. Kendall/Hunt. Dubuque, IA.

Morrison, A. and T. G. Andreadis. 1992. Larval population dynamics in a community of nearctic *Aedes* inhabiting a temporary vernal pool. Journal of the American Mosquito Control Association 8: 52-57.

O'Connor, C. T. 1959. The life history of *Mochlonyx cinctipes* (Coquillett) (Diptera: Culicidae). Annals of the Entomological Society of America 52: 519-523.

Pennak, R. W. 1989. Fresh-Water Invertebrates of the United States: Protozoa to Mollusca, 3rd. ed. John Wiley and Sons. New York.

Peters, R. H. 1983. The Ecological Implications of Body Size. Cambridge University Press. Cambridge.

Schneider, D. W. 1997. Predation and food web structure along a habitat duration gradient. Oecologia 110: 567-575.

Schneider, D. W. 1999. Snowmelt ponds in Wisconsin: Influence of hydroperiod on invertebrate community structure, p. 299-318. *In* [eds.], D.P.Batzer, R.B.Rader, and S.A.Wissinger, Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley and Sons. New York.

Schneider, D. W. and T. M. Frost. 1996. Habitat duration and community structure in temporary ponds. Journal of the North American Benthological Society 15: 64-86.

Schwartz, S. S. and P. D. N. Hebert. 1985. *Daphniopsis ephemeralis* sp.n. (Cladocera, Daphniidae): a new genus for North America. Canadian Journal of Zoology - Journal Canadien de Zoologie 63: 2689-2693.

Sih, A. 1987. Predators and prey lifestyles: an evolutionary and ecological overview, p. 203-224. *In* [eds.], W.C.Kerfoot and A.Sih, Predation: Direct and Indirect Impacts on Aquatic Communities. University Press of New England. Hanover, N.H.

• lusarczyk, M. 1995. Predator-induced diapause in *Daphnia*. Ecology 76: 1008-1013.

Taylor, B. E., R. A. Estes, J. H. K. Pechmann, and R. D. Semlitsch. 1988. Trophic relations in a temporary pond: Larval salamanders and their microinvertebrate prey. Canadian Journal of Zoology - Journal Canadien de Zoologie 66: 2191-2198.

Tilman, D. 1982. Resource Competition and Community Structure. Princeton University Press. Princeton, NJ.

Travis, J., W. H. Keen, and J. Juilianna. 1985. The role of relative body size in a predator-prey relationship between dragonfly naiads and larval anurans. Oikos 45: 59-65.

Wellborn, G. A., D. K. Skelly, and E. E. Werner. 1996. Mechanisms creating community structure across a freshwater habitat gradient. Annual Review of Ecology and Systematics 27: 337-363.

Westwood, A. R., G. A. Surgeoner, and B. V. Helson. 1983. Survival of spring *Aedes* spp mosquito (Diptera: Culicidae) larvae in ice-covered pools. Canadian Entomologist 115: 195-197.

Wiggins, G. B., R. J. Mackay, and I. Smith. 1980. Evolutionary and ecological strategies of animals in annual temporary pools. Archiv für Hydrobiologie, Supplement 58: 97-206.

Wilson, M. S. and H. C. Yeatman 1959. Free-living Copepoda, p. 735-861. *In* [eds.], W.T.Edmondson, H.B.Ward, and G.C.Whipple, Fresh-Water Biology. John Wiley and Sons. New York.

Young, A. M. 1967. Predation in the larvae of *Dytiscus marginalis* Linneaus. Pan-Pacific Entomologist 43: 113-117.

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

APPENDICES

APPENDIX A

Record of Deposition of Voucher Specimens*

The specimens listed on the following sheet(s) have been deposited in the named museum(s) as samples of those species or other taxa which were used in this research. Voucher recognition labels bearing the Voucher No. have been attached or included in fluid-preserved specimens.

Voucher No.: 2000-3

Title of thesis or dissertation (or other research projects):

Invertebrate Trophic Relationships in Temporary Woodland Ponds in Michigan

Museum(s) where deposited and abbreviations for table on following sheets:

Entomology Museum, Michigan State University (MSU)

Other Museums:

Investigator's Name (s) (typed)
Michael J. Higgins

Date 1 May 2000

*Reference: Yoshimoto, C. M. 1978. Voucher Specimens for Entomology in North America. Bull. Entomol. Soc. Amer. 24:141-42.

Deposit as follows:

Original:	Include as Appendix 1 in ribbon copy of thesis or dissertation.
Copies:	Included as Appendix 1 in copies of thesis or dissertation. Museum(s) files. Research project files.

This form is available from and the Voucher No. is assigned by the Curator, Michigan State University Entomology Museum.

APPENDIX B

Voucher Specimen Data

Page ____ of ____ Pages

		Number of:							
Species or other taxon	Label data for specimens collected or used and deposited	Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	Museum where deposited
Aedes stimulans (Walker)	Temporary pool, 1 km east of Lake Lansing, 11 May 1998 coll. Michael J Higgins		5			5	5		

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Michael J Higgins

Date 1 May 2000

Voucher No. 2000-3

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

5/1/2000

Curator Date