Fall 1996

Life history and reproductive biology of the estuarine nudibranch Tenellia adspersa (Nordmann, 1845)

Charles Morgan Chester

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LIFE HISTORY AND REPRODUCTIVE BIOLOGY OF THE
ESTUARINE NUDIBRANCH Tenellia adspersa (NORDMANN, 1845).

BY

Charles Morgan Chester
B.S. University of Rhode Island. 1986
M.S. University of Rhode Island. 1991

DISSERTATION

Submitted to the University of New Hampshire
in partial fulfillment of the
Requirements for the Degree of

Doctor of Philosophy

in

Zoology

September, 1996
This dissertation has been examined and approved.

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30 August 1996
Date
Acknowledgment

No one works in a vacuum, and this dissertation would not be in its present form without the help and support of many people. I would like to thank my advisor, Dr. Larry Harris for his enthusiasm, and support throughout this study, and for encouraging me to pursue what I found interesting. I would also like to thank my committee members, Dr. J. Haney, Dr. E. Tillinghast, Dr. M. Litvaitis and Dr. A. Kuzirian for their constructive comments, valuable insights, enthusiasm, and never being too busy to answer questions.

My deepest appreciation goes to my wife, Kathleen and to Louie, Bailey, Ella, Boris (Bobo) and the late, great Mr. Louis Biddles for their steadfast love and devotion as well as helping to maintain my sanity and reminding me of what is important in life.

I would like to thank graduate student friends past and present for their encouragement and constructive comments. In particular, Chris and Diane Hartleb, Scott (fish-bo) Orringer, Jon and Alexis Runstadler, Marc Simmons, Eric Lovely, Cheryl Gibeault, Sue Reidy, Rick Biche, Dave Carlon, Kadee Lawrence, Brad Peterson, and Dr. Ken Thomas (who only beat me by a few months).

Additional thanks to Saud Al-Ayoub, Dave DeCarle, Lauralynn Dyer, Sue Iskra, Scott Mulliken, Melisa Nyberg, Barbara Piel, Laura Rodriguez, Fred Rotman, Kate Sardi, Kim Seidl, and Christa Williams for their assistance in culturing both nudibranchs and spawn, and for keeping the lab a fun and exciting place.

I wish to thank the Town of Durham, W. and C. Buckley, the Great Bay Marine Inc., and the Portsmouth Fishing Co-op for allowing me to conduct the field on their properties. Support for the field work was provided by a Lerner-Gray Grant from the American Museum of Natural History. Portions of the laboratory work was supported by a Grant-in Aid of Research from Sigma Xi, a grant from Hawaiian Malacological Society and from the Center for Marine Biology, University of New Hampshire.
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ABSTRACT

LIFE HISTORY AND REPRODUCTIVE BIOLOGY OF THE ESTUARINE NUDIBRANCH Tenellia adspersa (NORDMANN, 1845).

BY

Charles Morgan Chester
University of New Hampshire, September, 1996

Tenellia adspersa is a small (5-7mm) estuarine nudibranch. Tenellia lives in a highly variable environment and feeds on many gymnoblastic and calyptoblastic hydroids. I undertook a field study at four sites in the Great Bay Estuary (Durham, New Hampshire) to assess the amount of variability in abiotic and biotic factors. Environmental parameters (temperature and salinity) and the availability of hydroid prey (Cordylophora lacustris and Obelia spp.) varied within the generation time of Tenellia and could affect its life history.

Laboratory studies compared the growth rate of the hydroid, Cordylophora to the growth rate and predation rate of Tenellia. The results suggested that Tenellia could exhaust its food supply within a single generation, depending on initial colony size. Demographic studies comparing nudibranch life history reared on two common hydroids (Cordylophora and Obelia) found that Tenellia could vary its life history (age and size at maturity) in response to differences in hydroid species.

Tenellia displays poecilogony (more than one developmental mode). Proximate environmental sources were investigated in an examination of the effect of adult nutritional state on reproductive output, offspring development, survival and growth. I compared nudibranchs provided with an ad libitum diet of Cordylophora to individuals starved up to four days. Development was plastic: eggs from the same spawn produced veligers that either hatched as non-feeding swimming larvae or metamorphosed within the capsule, hatching as benthic juveniles. This strategy was dependent on egg size, with eggs larger than 125 μm hatching as benthic juveniles. Adult nutritional state affected egg size and the frequency at which the two developmental modes were
expressed. Five to 20 percent of the eggs within a spawn hatched as benthic juveniles under well-fed conditions. In contrast, all of the eggs within a spawn hatched as swimming larvae and metamorphosed into smaller juveniles under starvation conditions. However, adult starvation did not affect juvenile growth and survival, suggesting that any cost associated with maternal investment were confined to the pre-metamorphic stages. Furthermore, these results suggested that the plastic developmental strategy employed by *Tenellia* may represent a bet-hedging strategy allowing *Tenellia* to survive and reproduce in an unpredictable estuarine environment.
GENERAL INTRODUCTION

All environments are temporally variable. The type, size and periodicity of the environmental change determines the nature of the heterogeneity (Reice, 1994). Random or irregular events, such as storms, may cause temporal fluctuations or more permanent changes. Daily, lunar, solar or seasonal cycles may cause regular oscillations, such as seasonal changes in temperature, photoperiod, or salinity, each having a different periodicity.

Estuarine environments display a complex set of periodically changing environmental factors that limit the colonization of these areas to organisms with a wide range of ecological adaptations (Casper, 1967). Stehr (1964) suggested that if the periodicity in environmental change falls within one generation then selection will favor stabilizing mechanisms for maintaining variability in life history traits. One such mechanism for maintaining variability is phenotypic plasticity. Phenotypic plasticity refers to multiple phenotypes produced by a single genotype and is determined by the interaction of the genotype with the environment (Caswell, 1983). In these changing environments, plasticity may be advantageous for an organism by allowing it to survive and reproduce in a wider range of habitats.

An examination of the life history strategies employed by organisms living in variable estuarine environments allows insights into mechanisms of survival and reproduction. Life history strategies are suites of traits that determine the survival and reproduction of an organism. These traits are co-adapted such that altering one trait may result in concomitant changes in other traits (Stearns, 1980). There are several critical events in a life cycle that have dramatic effects on an organism's abundance and distribution. This dissertation focuses on two of these events, age and size of reproductive maturation and the role of offspring investment.
Reproductive maturation is an important life history event because it is a costly process for an organism, involving a redirection of resources from somatic growth and maintenance to gonad growth, gonad development and gamete production (Bernardo, 1993). As a result, the age and size at which an organism matures can have profound effects on fitness across a wide range of life histories (Stearns, 1992). Age and size at maturation have been shown to vary among species, among populations within species and among individuals within a population (Bell, 1980; Stearns, 1983). Organisms generally display one of five patterns of age and size at maturity (Stearns and Koella, 1986). Organisms can: (1) mature earlier at a smaller size, (2) mature at an equal age but a smaller size, (3) mature later at a smaller size, (4) mature later at an equal size, or (5) mature later at a larger size.

Age and size at maturity represents a trade-off between survival and reproduction. Organisms that mature early may benefit from a shorter period spent as a juvenile, resulting in a higher survival at maturity (Bell, 1980) and a shorter generation time (Cole, 1954). Fecundity is related to body size (i.e., gonad) in many organisms (Stearns, 1992, Roff, 1992). Delaying maturity may allow for further growth and a subsequently higher initial fecundity. Delaying maturity may also reduce the instantaneous juvenile mortality rate of the subsequent generation by allowing a higher quality of offspring to be produced (Stearns, 1980). Later maturity may also allow for more reproductive events due to a longer life span, thus increasing lifetime fecundity (Stearns, 1980).

Phenotypic plasticity in maturation accounts for much of the variation observed in age and size at maturity (Stearns, 1980). Maturation is dependent on an individual's growth and mortality rates (Perrin and Rubin, 1990). Variation in growth and mortality rates will cause variation in the optimal age and size at which maturation takes place. The advantage of phenotypic plasticity for organisms living in spatially or temporally changing environments is that it allows an organism to survive in a wider
range of habitats. Natural selection should therefore favor genotypes that present adaptive phenotypic plasticity for age and size at maturity, particularly if the environments fluctuate more rapidly than the generation time of the organism (Perrin and Rubin, 1990; Bernardo, 1993).

Another critical period in organisms with complex life cycles is early development. Early development can have profound effects on adult distribution and abundance. Capinera (1979) suggested that variability in offspring size represents an expression of phenotypic variation allowing progeny in continually changing environments to cope with instabilities. Kaplan and Cooper (1984) proposed that if different egg sizes are favored under different environmental conditions that stochastically vary among generations, then within-individual variation in egg size could represent a bet-hedging strategy. Organisms using bet-hedging strategies sacrifice their optimal fitness to reduce the probability of complete failure (Cohen, 1966; Seger and Brockman, 1987). Crump (1981) found that in tree frogs the distribution of egg sizes in species that bred in temporary pools had a broader than normal distribution, suggesting a "bet-hedging" response to environmental variability. The distribution of egg sizes for species that bred in permanent ponds was narrower suggesting stabilizing selection around an optimal egg size. Hickman (1975, 1977) found variation in the energy allocated to seeds in the annual plant, Polygonum cascadense, in response to annual short-term unpredictability of the environment.

Development is the period from fertilization or egg laying to metamorphosis into a juvenile. It involves a complex process of cell division, growth and differentiation of a single fertilized egg into an immature juvenile (Villee, et al., 1978). Following the terminology of Giese and Pearse (1974), an embryo refers to all developmental stages occurring within the parent, egg case or egg membrane, whereas, a larva is a free swimming stage that passes through a metamorphosis to form an immature juvenile. Development may be divided into two phases of varying length,
(1) the embryonic phase, which is represented by the period from fertilization or egg laying to hatching, and (2) the larval phase, which is the period from hatching to metamorphosis.

Traditionally, invertebrate developmental modes have been differentiated based on the presence or absence of a larval phase (termed indirect or direct development, respectively) (Figure 1) (Thorson, 1950; Thompson, 1967; Vance, 1973; Bonar, 1978). Species may also be classified based on the probability of dispersal. Pelagic development refers to species with a larva that disperses in the water column, whereas, non-pelagic development refers to species with a larva that does not disperse in the water column. Species with a larval phase are further classified based on larval nutrition. Lecithotrophic development refers to species with larvae that rely entirely on yolk material within the egg for nutrition. Planktotrophic development refers to species with larvae that must supplement their nutrition by feeding on planktonic organisms to metamorphose.

With few exceptions, most species of opisthobranch mollusks fit into one of four modes: (1) planktotrophic development, (2) pelagic lecithotrophic development, (3) capsular metamorphic (or non-pelagic lecithotrophic), and (4) capsular ametamorphic development (or direct). Capsular metamorphic development refers to species in which a veliger is formed and metamorphoses within the egg capsule. Capsular ametamorphic development refers to species in which the veliger stage is suppressed or absent.

However, there are conspicuous and important exceptions. Thorson (1950) described planktotrophic larvae with a short pelagic phase that can feed in the plankton but can settle and metamorphose without feeding. These larvae appear to arise from larger eggs than those of the strict planktotrophic larvae. Additionally, some lecithotrophic larvae can facultatively feed in the plankton (e.g. *Phestilla sibogae* (Kempf and Hadfield, 1985) and *Adalaria proxima* (Kempf and Todd, 1989). This
feeding may significantly prolong the pelagic larval period. Further, it appears that some larvae can use dissolved organic material (DOM) as a food source (e.g. *Haliotis rufescens* (Jaeckle and Manahan, 1989)) (for review see Wright and Manahan, 1989; Manahan, 1990; Jaeckle and Manahan, 1992).

Thus, it appears that at least within the Mollusca, development does not fit into discrete modes. Development may be viewed as a continuum ranging from organisms with a short embryonic phase and a long larval phase (pelagic planktotrophic), to organisms with a moderate embryonic phase and a short larval phase (pelagic lecithotrophic), to organisms with a long embryonic phase and no larval phase (non-pelagic, direct, capsular metamorphic, capsular ametamorphic, etc.) (For review see Hadfield and Switzer-Dunlap, 1984).

Egg size is positively correlated with developmental time, developmental mode and energy investment (Thorson, 1946; 1950; Rice, 1950; Chia, 1976; Strathmann and Veddar, 1977; Hermans, 1979; Turner and Lawrence, 1979; Todd and Doyle, 1981; Hadfield and Switzer-Dunlap, 1984; Emlet et al., 1987). Assuming an organism has a finite amount of energy available for reproduction, then an organism that produces small eggs can produce more eggs compared to those that produce larger eggs (Thorson, 1950; Vance, 1973; Smith and Fretwell, 1974; Stearns, 1976; Bridges, 1993).

The main advantage conferred by a pelagic dispersal phase is the possibility of settling in new areas allowing genetic exchange over a large population, increasing geographic range and occupation of unstable and ephemeral habitats (Thorson, 1950; Mileikovsky, 1971; Vance, 1973; Crisp, 1974; Strathmann, 1974; Strathmann, 1985; Pechenik, 1990). However, a potential disadvantage of a pelagic larval period is the risk of mortality due to predation, or dispersal from suitable settlement sites (Thorson, 1950; Vance, 1974).
Planktotrophic larvae allow flexibility in the duration of their pelagic dispersal phase because the larvae are relying on an external food source as opposed to a finite endogenous reserve. This strategy may be disadvantageous in that these larvae are depending on a potentially patchy resource and may starve to death before settlement (Thorson, 1950; Vance, 1974; Olson and Olson, 1989).

Environmental factors, especially temperature and salinity, can have important consequences for development time. For most invertebrates, temperature has been shown to have a strong influence on embryonic time. For example, Todd and Doyle (1981) found that time to hatching in *Onchidoris bilamellata* decreased as temperature increased. Hagerman (1970) found that developmental time in *Elysia viridis* increased with decreasing salinity. Additionally, the percent mortality and percent abnormal development was higher at lower salinities. Harris et al. (1980) also found that development times in *Tenellia adspersa* increased with decreasing salinity. Clearly, fluctuations in temperature and salinity will be more important to organisms living in stressful environments, such as the intertidal zone (Newell, 1969) or in estuaries (Kinne, 1967).

In most opisthobranch species, egg size within and between individuals is comparatively invariant (Todd and Doyle, 1981), and developmental mode is generally considered rigid and species-specific (Bouchet, 1989). However, a few instances of switching of egg size and developmental mode have been observed (For review see Hoagland and Robertson, 1988). Developmental patterns vary geographically (e.g., Rasmussen, 1944; West et al., 1984), within a single population (e.g. Eyster, 1979) or seasonally (Clark and Goetzfried, 1978). Mechanisms explaining developmental variability in opisthobranch mollusks are generally lacking. Clark and Goetzfried (1978) proposed that an interruption of the adult's food supply or the utilization of a sub-optimal diet may induce a shift in developmental mode to one with a higher dispersal ability. This requires that gametogenesis continue despite nutritional...
insufficiency. Vitellogenesis would then decline or be prematurely terminated, resulting in smaller ova. Additionally, if hatching is controlled by an exhaustion of embryonic food reserves, then the smaller ova would hatch at an earlier stage. Another proposed mechanism is the result of variable degradation of the outer mucous layers of the spawn mass. The eggs of most opisthobranchs are enclosed in a gelatinous spawn mass that is attached to the benthos, and consists of three components: a nutritive albumen layer surrounding the eggs, an egg capsule enclosing eggs singly or in multiples, and a mucous layer called the egg matrix (Figure 2) (Ghiselin, 1963; Hadfield and Switzer-Dunlap, 1984). The capsule and matrix are both composed of mucopolysaccharides (Todd, 1981). The effect of bacteria, ciliates, nematodes, harpacticoid copepods, water movement and other extrinsic biotic and abiotic factors may be important in weakening this matrix and allowing escape (Hurst, 1967; Harris, 1973; Harris, 1975; Harris et al., 1980; Todd, 1981). In laboratory cultures, artificial delay of hatching has been observed in spawn masses maintained in static culture (no aeration, and no daily change of filtered seawater) compared to spawn masses maintained in flowing seawater (Davis, 1967; Hurst, 1967; Harris, 1975; Harris et al., 1980; Todd and Doyle, 1981). Eyster (1986) suggested that flowing water may provide more oxygen to developing embryos and/or may increase the rate of diffusion of embryonic wastes out of the capsule which otherwise may inhibit development. Harris (1973) and Harris et al. (1980) observed that veligers of *Phestilla sibogae*, *P. melanobrachia*, *Aeolidea papillosa* and *Tenellia adspersa* will not attempt to hatch from their capsules until the matrix has broken down sufficiently to release the capsules. Gibson and Chia (1989) found a diffusible compound present in the matrix that induces capsular metamorphosis in the cephalaspid *Haminoea callidegenita*. As the matrix erodes, less of the compound is present and the more likely it becomes that veligers will hatch out before metamorphosis.
Further, the breakdown of this matrix may be related to adult nutritional state. The egg matrix is secreted by the mucous gland (Ghiselin, 1966). An interruption in food supply or the utilization of a sub-optimal food may cause animals to secrete a weaker egg matrix that may erode at a faster rate, allowing veligers to hatch out at an earlier stage.

Tenellia adspersa is a small (5-7mm) aeolid nudibranch commonly found in New England estuarine environments. Tenellia feeds on a variety of gymnoblastic and calyptoblastic hydroids, including Obelia comisseralis, Campanularia flexulosa, Gonothyraea loveni, Ectopleura dumortieri, Bougainvillea carolinensis and Cordylophora lacustris (Clark, 1975). Most of these hydroids are seasonally abundant in the Great Bay Estuary (New Hampshire), and can be easily cultured in the laboratory, thus providing Tenellia with a potentially diverse diet.

Tenellia adspersa is a subannual species with a generation time of 20 days from egg to egg at 30°/oo salinity when raised on Cordylophora and a maximum age of 30-40 days (Harris et al., 1980). Development typically takes place in 6-7 days at 20° C. Competent veligers hatch out and undergo metamorphosis within 24 hours if hydroids are available. Juvenile growth is exponential with maturity occurring about 10 days afterwards at a size of about 4.5 mm in length. Growth rate then decreases with a maximum size of 6-7 mm.

Tenellia is a simultaneous hermaphrodite with internal fertilization and a triaulic reproductive system (Chambers, 1934). Tenellia lays 3-4 spawn masses per day for approximately 15 days. Each spawn mass contains an average of 30 eggs, each surrounded by its own egg capsule. Developmental mode has been considered pelagic lecithotrophic (Clark, 1975). However, capsular metamorphic development has been observed in this species (Harris et al., 1980; personal observation), and in the conspecific Tenellia pallida (Rasmussen, 1944; Eyster, 1979).
Due to the unpredictable nature of the estuarine environment in which *Tenellia adpersa* lives, its life history strategy is phenotypically plastic. This dissertation presents evidence in support of this hypothesis.

Chapter One describes patterns of change in the biotic and abiotic factors in the nudibranch's environment that *Tenellia* must cope with if it is to survive and reproduce. The results demonstrate that environmental parameters (i.e., temperature and salinity) vary within the generation time of the nudibranch. More interestingly, the abundance and distribution of hydroid prey also vary within the generation time and therefore may affect the life history of this organism.

Chapter Two compares the growth rate of the hydroid, *Cordylophora* (a major prey species) to the growth rate and predation rate of the nudibranch, *Tenellia*. This chapter suggests that *Tenellia* could exhaust its food supply within a single generation, depending on initial colony size.

Chapter Three compares the life history of *Tenellia* on three common hydroid species (*Cordylophora, Obelia* and *Hydractinia*). The results demonstrate that *Tenellia* varies its life history (age and size at maturity) in response to differences in hydroid species.

Chapter Four addresses plasticity at a specific point (early development) in the life history and looks at the effect of adult nutritional state on egg size and offspring development. This chapter demonstrates that *Tenellia* has a plastic developmental strategy that varies in response to adult nutritional regime.

Chapter Five investigates the effects of plasticity in offspring development on offspring life history with the intent of determining how closely related egg size is to parental investment and offspring development.

Chapter Six deals further with the variability in parental allocation by investigating the effect of variation in maternal investment on offspring growth and survival. The results indicate that although egg size and offspring development varies
in response to adult nutritional state, juvenile growth and survival do not. This suggests that any costs to the offspring resulting from changes in egg size are confined to pre-metamorphic stages.
Figure 1. Generalized invertebrate life cycle illustrating the various developmental modes. Modified after McEdward and Janies (1993).
Figure 2. The anatomy of a nudibranch spawn mass.
CHAPTER I

RECRUITMENT AND EARLY COMMUNITY SUCCESSION OF FOULING ORGANISMS WITHIN THE GREAT BAY ESTUARY

Introduction

One of the basic questions in ecology is how does the composition and structure of communities vary in time and space. Succession is a sequential change in community composition and structure over time (Connell and Slatyer, 1977; Drury and Nisbet, 1973; Horn, 1974; Miles, 1987; Pickett, 1976). Connell and Slatyer (1977) proposed that succession takes place by one of three mechanisms, described as the facilitation, inhibition and tolerance model, respectively. Under the facilitation model, early successional species facilitate the recruitment and establishment of later arrivals. Under the inhibition model, early successional species inhibit the recruitment and establishment of later species. Finally, under the tolerance model, early successional species have no effect on the recruitment and establishment of later species.

Studies in the rocky intertidal zone have been instrumental in determining the role and relative importance of predators in structuring communities. For example, the presence of keystone predators may allow competitively inferior species to persist and allow higher species diversity by preying on the competitive dominant in a system (Paine, 1966; Paine, 1969). Other studies in the rocky intertidal zone (See for example: Lubchenco, 1978; Marsh, 1986; Menge, 1976; Menge, 1978; Menge, 1991; Menge and Sutherland, 1976) have further demonstrated the importance of predation in structuring these communities. Studies on sea urchins in the rocky subtidal have demonstrated their importance in preventing kelps and other fleshy algae from
dominating the system (Martin, et al., 1988 and references therein).

Although the role of predation in structuring communities has been well studied, its role as a mechanism for successional change has been largely ignored (Connell and Slatyer, 1977). Recent studies, involving the manipulation of predators, indicate that interactions between early successional species and their predators are likely to be important in determining the rate of succession (Reviewed in Hawkins and Hartnoll, 1983). These predators may also alter the successional sequence by removing or reducing the abundance of an early colonist, allowing another species to become established. Farrell (1981) predicted that predators will either slow, accelerate, or have no effect on the speed at which succession takes place depending on how early colonists affect the recruitment of later species (i.e., facilitation, inhibition or tolerance).

Hydroids are typical early colonists in fouling communities (Clark, 1975). They often have ephemeral life history and distribution patterns, and tend to be succeeded by other species such as barnacles, tunicates and mussels (Clark, 1975; Haderlie, 1969; Harris and Irons, 1982; Scheer, 1945). Nevertheless, hydroids appear to play an important role in determining the kinds of species that recruit and succeed them. Hydroids have been shown to facilitate mussel and tunicate recruitment while inhibiting barnacle recruitment (Dean, 1981; Dean and Hurd, 1980; Standing, 1976). If not removed by predation, some hydroids (i.e., Obelia and Hydractinia) have been shown to effectively hold space for long periods of time (months to years) (Standing, 1976; Karlson, 1978).

Members of the molluscan Suborder Aeolidacea are colorful shell-less gastropods, most of which are important partial predators on colonial cnidarians such as hydroids, octocorallian and scleractinian corals (Todd, 1981; Todd, 1983). Predation by aeolids plays a significant part in structuring hydroid communities by creating physical gaps in prey colonies, altering the population structure of the prey, or inducing changes in the prey's growth form (Harris, 1987). In low numbers, aeolids may have a
limited impact on a hydroid community, because nudibranch feeding rates are arithmetic while hydroid colonial growth is exponential. Such limited predation by nudibranchs may actually improve a hydroid's persistence by inducing growth form changes in a colony (e.g., Gaulin, et al., 1986). In addition, hydroids may have temporal or spatial refuges from their nudibranch predators. Hydroids may also escape in time by reproducing rapidly enough to prevent the nudibranch population from limiting their increase. However, at higher abundances, the impact of aeolids on hydroids is more substantial, and may ultimately lead to the removal of the hydroid colony. As a result of the aeolid's feeding behavior, portions of the hydroid's perisarc may remain behind after the hydroid has been eaten, thus modifying the substratum and potentially altering the course of succession.

Estuarine environments are intrinsically variable on both spatial and temporal scales. Because environmental factors within estuaries are highly variable and often stressful, colonization is limited to organisms with a wide range of ecological adaptations (Casper, 1967). This pattern will add further complexity to successional patterns observed.

A number of aeolid species have adopted a life history of short cycles coupled with high fecundity and a fairly long planktonic larval phase to take advantage of temporally unpredictable but often abundant food resources. This life history pattern can have significant consequences for the hydroid prey and potentially determine the persistence of a hydroid colony. This chapter investigates the importance and role of predation in communities undergoing successional change by observing the relationship between hydroids, their predators and other organisms inhabiting the fouling community at a series of sites within an estuary.
Materials and Methods

Due to the ephemeral nature of the hydroid populations, variation in recruitment and changes in species abundances are likely to occur rapidly. These changes were monitored on plexiglass panels exposed for up to a month from June to November of 1993. Plexiglass panels (13 x 8 cm) were suspended in four replicate wooden arrays that were deployed at each of four stations within the Great Bay Estuary (Figure 1.1). The panels were held vertically to remove any potential effects of sedimentation on recruitment (e.g., Harris and Irons, 1982) and were allowed to rotate in the current so that both sides received approximately equal current regimes (Figure 1.2A). The wooden array shaded the panels, inhibiting algal growth. Each set of four panels on a single array was numbered 1 through 4 corresponding to the number of weeks that it was exposed before being removed and replaced. Each week, at low tide, panels were removed according to their specific schedule (Figure 1.2B) and placed in a bag. Removed panels were replaced with the exception of the three week panels. These were removed after three weeks and new panels put in the following week. Salinity and temperature at each site was recorded at the same time as the panels were replaced.

Replaced panels were returned to the lab where all organisms larger than 1 mm were identified to the lowest taxonomic level using local faunal keys (Frazer, 1944; Smith, 1964). The total number of individuals or colonies were recorded, and the percent cover of each non-motile species was measured using a grid of 100 randomly distributed points.

Statistical analysis was performed using SYSTAT (Systat Inc., Evanston, IL.). Temporal variation in recruitment and information on the abundance and distribution of hydroids and nudibranchs was investigated by comparing panels exposed for the same amount of time (e.g. comparing the one week panels). Relationships between organisms during the early successional sequence and information on the persistence of
both hydroids and their effect on the successional process was investigated by comparing panels exposed for one, two, three and four weeks with each other. Pearson correlation coefficients were used to study relationships between organisms inhabiting the panels. Means and standard errors are used throughout.

Results

Temperature and Salinity.

The four stations differed in the temperature and salinity regimes in which they were exposed (Figure 1.3). At station 1, salinity was highest (29.2 ± 1.2 °/oo salinity, n = 21, range 27-32) and temperature lowest (14.4 ± 3.3 °C, n = 21, range 8-18) with very little fluctuation in either. At station 2, salinity was generally lower (27.8 ± 1.3 °/oo salinity, n = 21, 25-30) and temperature higher (16.4 ± 3.8 °C, n = 21, range 9-21). At station 3, salinity was lower still and more variable (23.7 ± 5.8 °/oo salinity, n = 21, range 0-28). Note that salinity reached zero on one sampling date. Temperature at this station was higher and also more variable (19.2 ± 5.5 °C, n = 21, range 8-27). Salinity was lowest at station 4 and highly variable (13.2 ± 8.1 °/oo salinity, n = 21, 0-24). Salinity was below 10 °/oo salinity on several sampling dates and was zero on two dates. Temperature was the highest and also more variable (20.1 ± 5.9 °C, n = 21, range 8-28).

Recruitment.

Forty-three species of invertebrates were found on the plexiglass panels (Figures 1.4 and 1.5). Most of these species were found at stations 1 to 3 (60% of the species were found at station 1, 56% at station 2 and 47% at station 3). A smaller number of species were found at station 4 (21% of the species found). Approximately half of these were colonial organisms (Figure 1.4). Recruitment patterns of marine invertebrates on to panels exposed for 2 weeks and longer was the same as on panels exposed for 1 week.
A number of cnidarians recruited to the fouling panels, the majority of which were gymnoblastic and calyptoblastic hydroids. One individual of the anemone *Haliplanella luciae* was found on panels at station 2 in August. The gymnoblastic hydroids that appeared on the panels were *Tubularia crocea*, *T. larynx*, *T. indivisa* and *Eudendrium* sp.. *Eudendrium* sp. was only found at station 1 in September while *Tubularia* spp. was found in large numbers at stations 1 and 2 and in small numbers at station 3 during July. There was no recruitment of *Tubularia* spp. at station 4. At station 1, *Tubularia* recruitment occurred from June through October with peak recruitment in July and early August (July: 11.5 ± 2.9 colonies/100cm², n = 4; August: 9.3 ± 6.6 colonies/100 cm², n = 4) (Figure 1.6A). Recruitment at station 2 followed a similar pattern but was an order of magnitude less and peak recruitment occurred later (middle to late August).

A variety of small calyptoblastic hydroids appeared on the panels from June through October. These included *Obelia commiseralis*, *Campanularia flexulosa*, *C. gelatinosa* and *Gonothraea loveni*. Due to difficulties in identification, these hydroids were grouped together and called campanularid hydroids. Campanularid recruitment occurred at stations 1, 2, and 3 during June and July, with the highest recruitment occurring in the first few weeks of June (Figure 1.6B). A few colonies were found sporadically at stations 1, 2 and 3 in August, September and October.

Another hydroid, *Cordylophora lacustris*, was conspicuously absent from the study. A small colony of *Cordylophora* was found in June a little upriver from station 4, but had disappeared in the following months.

The bivalve, *Mytilus edulis*, recruited in low numbers (2-3/100 cm²) at station 1 from June to September, with a pulse (8.25 ± 4.03/100 cm², n = 4) occurring during the second week in July (Figure 1.7). This pulse coinciding with the peak in *Tubularia* recruitment. Recruitment was patchy at station 2 and peak recruitment occurred towards the end of August. Recruitment did not occur at either stations 3 or 4.
The barnacle, *Balanus improvisus* recruited in very low numbers, primarily to stations 3 and 4 from June to October (Figure 1.8). Recruitment at station 4 was highest in the end of June and beginning of July (54.0 ± 29.7, n = 4) and then declined. Recruitment at station 3 peaked later in the season, reaching the highest abundance the end of August (6.5 ± 1.9, n = 4).

Colonial tunicates, consisting of *Botryllus schlosseri*, *Botrylloides diegensis* and *Diplosoma macdonaldi*, appeared on the panels at station 1 in every month except June (Figure 1.9A). Peak recruitment occurred in the middle of August (8.3 ± 1.2, n = 4). Recruitment was sporadic at station 2 and appeared to peak later in September and October. No recruitment of colonial tunicates occurred at stations 3 and 4. The only solitary tunicate, *Mogula* sp., recruited in very low numbers at stations 2, 3 and 4 from July to September.

Encrusting bryozoans consisted of *Electra crustutenta*, *Membranipora membranacea*, *Hiplo diplosa* sp., *Aeverillia armata*, *Scruparia ambigua*, and *Cribilina punctata*. Recruitment occurred in very low numbers at all stations in every month (Figure 1.9B), with a peak recruitment at station 3 and 4 occurring at the end of July and beginning of August. Some recruitment occurred at station 1 in September.

The polychaete, *Spirobis spirillum*, recruited in very low numbers at station 1 at the end of August and beginning of September (August 31: 0.25 ± 0.25 individuals/100cm², n = 4; September 7: 0.50 ± 0.50 individuals/100cm², n = 4). No recruitment occurred at stations 2, 3 or 4.

**Relations between Hydroids and Other Organisms.**

Comparisons of *Mytilus* and hydroid abundances revealed significant positive correlations at station one during the months of June and July (Table 1.1). A positive correlation between mussels and campanularid hydroids was observed at station 1 in June (Figure 1.10A). Positive correlations between mussels and *Tubularia* were also observed at station 1 during June and July (Figures 1.10B and C).
There were no correlations between the barnacle, *Balanus improvisus* and *Tubularia* spp. at any station, during any months (Table 1.2). However, barnacle abundance was positively related to campanularid abundances at station 3 during August and September. This was at a time when the abundance of campanularid hydroids was relatively low.

Significant correlations between colonial tunicates and *Tubularia* spp. occurred at station 1 during July and September and at station 2 during August (Table 1.3). No relationship was found between tunicates and campanularid hydroids. Encrusting bryozoans, and *Spirorbis* recruited in numbers that were too low to allow comparisons with the abundance of hydroids.

**Predator/Prey Relationships.**

**Tubularia Predators.**

Predators of *Tubularia* spp. that were found on the plexiglass panels included the nudibranch *Dendronotus frondosus* and the pycnogonid *Phoxichilidium tubulariae*. Both predators were only found at station 1. *Dendronotus* was found on these panels during July when *Tubularia* abundance was highest. Large numbers of *Phoxichilidium* were also found in July and a few were found in September. At station 1, during the month of July, the abundance of *Tubularia* increased with exposure. It reached a plateau of approximately 50% cover after three weeks (Figure 1.11). Predators also increased and reached approximately 7 individuals per 100 cm$^2$ at the end of four weeks.

**Campanularid Predators.**

Predators on campanularid hydroids included the nudibranchs *Tergipes tergipes*, *Eubranchus exiguum*, and *Tenellia adspersa*. *Tergipes* and *Eubranchus* were found in abundance during June only at stations 1 and 2 and were not found during later months. *Tenellia adspersa* was found in abundance at station 1 during June, and at much lower densities at Stations 1, 2 and 3 during July and August.
During June, the abundances of predator and prey varied (Figure 1.12). At station 1 the percent cover of campanularid hydroids, *Tenellia* and nudibranclh spawn masses all increased with exposure. At station 2 the numbers of nudibranchs and spawn masses increased while the percent cover of hydroids increased and reached a plateau after three weeks. At station 3 all three increased in the first three weeks and then decreased in the last week. Campanularid hydroids actually reached zero by week 4. A visual inspection of the 4 week plates revealed that the perisarc of the stolons remained but no living hydroids were found. The presence of spawn masses appearing beginning week 4 at stations 1 and 2 and beginning week 2 at station 3 suggest a generation time of 2 to 4 weeks which correlates well with observed generation times for *Tenellia* raised on *Obelia* (Chapter two).

**Discussion**

Three basic recruitment patterns were observed in the present study for the marine invertebrates in the Great Bay Estuary with respect to station and time. One pattern was a peak in recruitment earlier at station 1 and later at stations 2-4. This pattern was observed for *Tubularia* spp., *Mytilus edulis*, and tunicates. A second pattern was earlier recruitment at stations within the Great Bay (station 3 and 4) and later recruitment at the mouth of Portsmouth Harbor (station 1). This was observed for *Balanus* and the byrozoan species found. A third pattern observed for the campanularid hydroids was peaks occurring at each station (except station 4) at the same time.

The recruitment of hydroids to the fouling panels was highly variable. Campanularid hydroids recruited to station 1-3 primarily in the spring. However, colonies were found sporadically at these three stations from July through October, demonstrating that campanularid hydroids were present and available to recruit during the entire study period. The recruitment patterns also suggest that hydroid abundance and distribution patterns were patchy, temporally unpredictable and could change in as
short a time as a week. *Cordylophora lacustris* displayed a different pattern. *Cordylophora* is only found in low salinity riverine systems and does not typically occur within the Great Bay itself (personal observations). Unlike the campanularid hydroids, *Cordylophora* appears to exist in a refuge of lower salinity where nudibranch predators cannot normally reach.

This recruitment pattern has implications for the life histories of the nudibranch predators that feed on campanularid hydroids. Todd (1983) proposed that nudibranchs preying on temporally and spatially unpredictable resources should have subannual life history strategies with an emphasis on rapid location, colonization, reproduction and dispersal. The three nudibranchs found (*Tergipes tergipes*, *Eubranchus* spp. and *Tenellia adspersa*) follow this pattern. All three are small (<10 mm maximum length) with short subannual life cycles (months) and high fecundities (Clark, 1975; Personal Observation). Both *Tergipes* and *Eubranchus* are opportunistic specialists on campanularid hydroids (Clark, 1975; Todd, 1981) and have planktotrophic larvae with a fairly long pelagic phase (month or more). *Tenellia* preys on a wide variety of calyptoblastic and gymnoblastic hydroids (Clark, 1975) and is unique in that it has a short pelagic lecithotrophic larval phase (Chapter 3). In addition, *Tenellia* was the only nudibranch species found higher in the estuary where salinities were lower.

In contrast, *Tubularia* spp. recruited only to stations 1 and 2 in mid-summer and was present in every month of the sampling. Predators on *Tubularia* also increased in abundance over time. However, there appeared to be no effect of predation on *Tubularia* colony abundance. This may have been the result of the panels being exposed for too short a time to detect an effect. It is also possible that predators do not have a large effect on *Tubularia* populations and some other factor (e.g., seasonal changes in abiotic factors) controls *Tubularia* abundances. Hughes (1983) found high summer mortality of *Tubularia* spp. due to the nudibranch, *Dendronotus frondosus*. Clark (1975) also observed high mortality of *Tubularia* spp. colonies during population...
peaks of the nudibranch, *Catriona aurantia*. However, when Cooper (1980) simulated nudibranch predation on *Tubularia crocea* colonies by clipping polyps, he found no difference in abundance between clipped and control colonies. In addition, Lovely (1995) did not observe a clear relationship between the abundance of *Tubularia* spp. colonies and predator abundance and concluded that the seasonal decline in *Tubularia* spp. was due to changes in environmental conditions.

*Mytilus edulis* recruited during mid-summer and peaked when *Tubularia* recruitment was highest. Positive correlations between hydroids (early colonists) and mussels (later colonists) were observed for both the large gymnoblastic hydroids (i.e., *Tubularia* spp.) and the smaller calyptoblastic hydroids (i.e., campanularid hydroids). This suggests that hydroids are facilitating mussel recruitment. Primary settlement of early plantigrades of *Mytilus edulis* preferentially occurs in substrates having filamentous or pitted structures (Bayne, 1964; Seed, 1969; Dean and Hurd, 1980; Okamura, 1986). Due to the filamentous nature of the hydroid perisarc, hydroids could be providing the preferred physical structure for the settling mussels. This facilitation has been demonstrated by other researchers. For example, Okamura (1986) found that the hydroid *Tubularia crocea* and the barnacle *Balanus improvisus* facilitated mussel recruitment in fouling communities. Dean and Hurd (1980) also found facilitation of mussel recruitment in a hydroid-tunicate assemblage (*T. crocea* and *Molgula manhattensis*).

If hydroids are facilitating the recruitment of organisms such as mussels, then nudibranchs may have an indirect effect on mussel recruitment by removing portions of the hydroid colony. Evidence of campanularid hydroid removal by nudibranchs was observed at some stations in this study, but not for the gymnoblastic hydroids. Such an indirect effect of predation on campanularid hydroids would weaken or remove the effect of mussel facilitation. In fact, the only significant relationship between campanularid hydroids and mussels occurred in June at station 1. This relationship was
weaker than the one observed between *Tubularia* spp. and mussels at the same time period. Van Tamelen (1987) demonstrated that molluscan herbivores could indirectly facilitate barnacle recruitment by removing the algae that was inhibiting barnacle recruitment.

In summary, this study suggests that hydroids are important early colonists in fouling communities that may facilitate recruitment of other organisms such as mussels. Nudibranchs are important predators that have the potential of rapidly removing their hydroid prey. This in turn may have indirect effects on the recruitment of other community members. In addition, this study demonstrated that calyptoblastic hydroids were temporally and spatially unpredictable in the Great Bay Estuary. All of the nudibranch predators that selectively prey on these hydroids have a subannual life histories that allows them to exploit this unpredictable resource.
Table 1.1. Pearson correlation coefficients relating the abundance of hydroids (*Tubularia* spp. and *Campanularid*) to the abundance of *Mytilus edulis* for three stations, during five months. Asterix = $P < 0.0001$, ns = not significant, NR = no results.

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<td>0.98 (*)</td>
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<td>NR</td>
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<td>0.89 (*)</td>
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<td>0.03 (ns)</td>
<td>0.17 (ns)</td>
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<td>0.26 (ns)</td>
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<td>August</td>
<td>0.10 (ns)</td>
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<td>0.06 (ns)</td>
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<td>September</td>
<td>0.28 (ns)</td>
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Table 1.2 Pearson correlation coefficients relating the abundance of hydroids (*Tubularia* spp. and campanularid) to the abundance of the barnacle, *Balanus improvisus* for three stations, during five months. Asterisks = $p < 0.05$, ns = not significant, NR = no results.

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<td><em>Tubularia</em></td>
<td>Campanularid</td>
</tr>
<tr>
<td>June</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>July</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>August</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>September</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>October</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
</tbody>
</table>

To ON
Table 1.3 Pearson correlation coefficients relating the abundance of hydroids (*Tubularia* spp. and campanularid) to the abundance of colonial tunicates for three stations, during five months. Asterisks = p < 0.05, ns = not significant, NR = no results.

<table>
<thead>
<tr>
<th></th>
<th>Station One</th>
<th></th>
<th>Station Two</th>
<th></th>
<th>Station Three</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><em>Tubularia</em></td>
<td></td>
<td></td>
<td><em>Tubularia</em></td>
<td>Campanularid</td>
</tr>
<tr>
<td>June</td>
<td>NR</td>
<td>NR</td>
<td></td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>July</td>
<td>0.50 (*)</td>
<td>NR</td>
<td></td>
<td>NR</td>
<td>NR</td>
<td>0.16 (ns)</td>
</tr>
<tr>
<td>August</td>
<td>0.33 (ns)</td>
<td>NR</td>
<td>0.75 (*)</td>
<td>NR</td>
<td>NR</td>
<td>0.80 (*)</td>
</tr>
<tr>
<td>September</td>
<td>0.45 (*)</td>
<td>NR</td>
<td>-0.05 (ns)</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>October</td>
<td>0.44 (ns)</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
</tbody>
</table>
Figure 1.1. Map of the Great Bay Estuary showing stations where the fouling arrays were deployed.
Figure 1.2. Diagram of fouling panel array (A) and deployment schedule (B).
Figure 1.3. Temperature and salinity at each station for each sampling date.

- **Salinity (°/oo)**

- **Temperature (°C)**
<table>
<thead>
<tr>
<th>Station</th>
<th>One</th>
<th>Two</th>
<th>Three</th>
<th>Four</th>
</tr>
</thead>
<tbody>
<tr>
<td>Month</td>
<td>JJASO</td>
<td>JJASO</td>
<td>JJASO</td>
<td>JJASO</td>
</tr>
</tbody>
</table>

Kingdom Protista
- *Zoothamnium* spp.

Kingdom Animalia
Phylum Cnidaria
- *Tubularia* spp.
- *Obelia commiseralis*
- *Campanularia* spp.
- *Gonothraea loveni*
- *Schizotricha tenella*
- *Haliplanella luciae*

Phylum Ectoprocta (Bryozoa)
- *Membranipora membranacea*
- *Hiplodiplosa* spp.
- *Electra crustutenta*
- *Aeverillia armata*
- *Scruparia ambigu*
- *Cribrilina punctata*

Phylum Entoprocta
- *Bartensia laxa*

Phylum Chordata
- *Mogula* spp.
- *Botrylloides diegenes*
- *Botryllus schlosseri*
- *Diplosoma macdonaldii*

Total Number of Species: 43683 44941 56651 33411

Percent Cover
- >50% ■
- 10-49% □
- 1-9% □
- 0% □

Figure 1.4. Distribution of colonial invertebrates at four stations within the Great Bay Estuary over a period of 5 months in the summer of 1993. Abundance data taken from panels exposed for 4 weeks. Total number of species is number of species found at a particular station, during a particular month.
Figure 1.5. Distribution of solitary invertebrates at four stations within the Great Bay Estuary over a period of 5 months in the summer of 1993. Abundances taken from panels exposed for 4 weeks. Total number of species is number of species found at a particular station, during a particular month.
Figure 1.6. Recruitment of *Tubularia* spp. colonies (A) and campanularid hydroids (B) to plexiglass panels at 3 stations in the Great Bay Estuary. No recruitment occurred at station 4.
Figure 1.7. Recruitment of *Mytilus edulis* to plexiglass panels at 3 stations in the Great Bay Estuary. No recruitment occurred at station 4.
Figure 1.8. Recruitment of *Balanus improvisus* to plexiglass panels in the Great Bay Estuary. Numbers are based on panels exposed for four weeks time.
Figure 1.9. Recruitment of colonial tunicates (A) and encrusting bryozoans (B) to plexiglass panels in the Great Bay Estuary. Tunicate species included *Botryllus schlosseri*, *Botrylloides diegensis* and *Diplosoma macdonaldi*. Bryozoan species included *Electra crustulenta*, *Membranipora membranacea*, *Hiplodiplosa* sp., *Scruparia ambigu*, and *Cribilina punctata*.

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Figure 1.10. Relationship between *Mytilus edulis* and campanularid hydroids (A), *Tubularia* spp. during June (B) and *Tubularia* spp. during July (C). All relationships occurred at station one.
Figure 1.11. The abundance of *Tubularia* spp. and its predators at station one during the month of July.
Figure 1.12. The abundance of campanularid hydroids and their predators at stations one, two and three during the month of June. Campanularid hydroids includes Obelia commiseralis, Campanularia flexulosa, C. gelatinosa and Gonothraea loveni.
CHAPTER II

LIFE HISTORIES OF A HYDROID/NUDIBRANCH ASSOCIATION

Introduction

The general paradigm for early community succession is that early colonists do not replace themselves (Connell, 1975). Connell (1978) suggested that succession could vary depending on factors such as disturbance, competitive interactions, and temporal availability of propagules. Connell and Slatyer (1977) proposed that established species could inhibit, facilitate or remain neutral in the recruitment of later successional stage species.

Hydroids are typical early colonists in fouling communities that tend to be succeeded by other species such as barnacles, tunicates and mussels (Harris and Irons, 1982). Hydroids often have ephemeral life history and distribution patterns, but can be important in affecting the recruitment of later successional species (Chapter One).

The majority of aeolid nudibranchs are partial predators, consuming portions of hydroid colonies (Todd, 1981; Todd, 1983). Aeolid nudibranchs may play significant role in structuring hydroid communities (Harris, 1987). Aeolid predation may create physical gaps in prey colonies, alter the population structure, or cause changes in the prey’s growth form. The impact of nudibranchs on hydroid colonies varies in relation to the number of predators in the colony. In low numbers, the impact may be limited because the hydroid colony can grow faster than the nudibranch predation rate. However, at higher abundances, the impact is more substantial and will ultimately lead to the removal of the hydroid colony.

A number of aeolid species have life histories with short generation times and high reproductive output to take advantage of temporally unpredictable, but often
abundant food resources. The majority of these species have an obligate planktotrophic larva that remains in the plankton for weeks to months. At least one of them (*Tenellia adspersa*) has non-pelagic lecithotrophic larvae that metamorphose within the egg capsule (Chapter 4). This has important implications for nudibranch population growth within a hydroid colony and for the persistence of that colony. For populations of aeolids having pelagic larval stages, population growth in a hydroid colony will be determined by settlement and metamorphosis of larvae from the plankton. For aeolids having capsular development (or non-pelagic lecithotrophic development), population growth following initial recruitment, will be determined by growth and reproductive rates of resident individuals within a hydroid colony. In the latter case, the aeolid’s populations will increase at an exponential rate, inundating the hydroid colony and consuming it in a relatively short time.

*Tenellia adspersa* is a small (5-7 mm) aeolid nudibranch commonly found in New England estuarine environments (Clark, 1975). *Tenellia* is a generalist that feeds on a wide variety of gymnoblastic and calyptoblastic hydroids. The majority of these hydroids are seasonally abundant within the Great Bay Estuary, New Hampshire (70°55'N, 43°05'W), existing as colonies on piers, floating docks, eel grass blades and other natural and artificial structures. Within the estuary, the distribution and abundance of these hydroids can change within as little as one week's time (Chapter 1). *Tenellia* has a plastic developmental mode where both pelagic lecithotrophic larvae and benthic juveniles are produced in the same spawn mass and by the same individual (Chapter 4). The present study explores the implications of nudibranch life history on the persistence of a hydroid colony.

**Materials and Methods**

The hydroid, *Cordylophora lacustris* was collected in September 1991 from a floating dock on the Oyster River, Durham, New Hampshire (70°55'N, 43°08'W) and was cultured on glass slides that were suspended in aquaria at 25°C and at a salinity of 41.
A small portion of a colony (~3 polyps) was placed under a monofilament line that was tightly wrapped around the slide. Colonies were fed nauplii of *Artemia* sp. daily.

The nudibranch, *Tenellia adspersa*, was collected in October 1991 in *Cordylophora* colonies that were on a floating dock on the Squamscott River, Rockingham, New Hampshire (70°56'N, 43°02'W) in the fall of 1991. Stock cultures were established in fingerbowls at 25°C and 25 °/oo and provided with an *ad libitum* amount of *Cordylophora*.

To compare nudibranch and hydroid life histories the following observations were gathered (1) measurement of hydroid colony growth, (2) determination of nudibranch life history and (3) measurement of predation rates on hydroid colonies. These data were used to construct a simple model of the affect of predation on the growth of a single hydroid stolon.

**Hydroid Colony Growth.**

Small pieces of *Cordylophora* containing approximately 3 polyps were attached to glass slides and suspended in aquaria at 4 salinities: 10, 15, 20 and 25 °/oo, with 10 replicates per salinity. These colonies were fed *Artemia* nauplii every other day. The number of polyps were counted every few days, and each colony was mapped at 60x using a Wild dissecting microscope equipped with a drawing tube. Stolon length was measured with a map measurer and the distance converted to nearest 0.1 mm. In *Cordylophora*, stolons and uprights grow at a fairly constant length (Fulton, 1963). Therefore, the inside diameter of stolon was measured to the nearest 1 μm from histological cross-sections with a compound microscope equipped with an ocular micrometer (diameter = 0.294 ± 0.002 mm, n = 10), and this value was used to calculate volume of stolon. Colonies were also cultured at 0, 5 and 30 °/oo as above, except colonies were not mapped, only number of polyps counted.
Nudibranch Life History.

Five newly laid spawn masses from *Tenellia* were haphazardly collected from the stock culture and raised with an *ad libitum* amount of *Cordylophora* in the same conditions as the stock culture. Water was changed daily and the number of nudibranchs recorded. Daily observations were made on nudibranch body length, number of spawn laid, number of eggs per spawn, and egg diameter with a binocular dissecting microscope equipped with an ocular micrometer.

Nudibranch Feeding Rates.

In order to measure feeding rates, nudibranchs of varying sizes were placed in colonies of *Cordylophora* and followed over time. The number of polyps were counted daily and the amount of living stolon measured as with the hydroid colony growth.

Hydroid Growth Rate

The number of polyps in a colony increase exponentially (Fulton, 1960; Fulton, 1961) while the growth of a part of a colony (i.e., one upright) is linear (Fulton, 1963). I chose to model the effect of a single nudibranch predator on both the number of polyps and on the volume of stolon tissue of a single growing stolon of *Cordylophora* through time. Polyp growth was calculated as

\[ P_{t+1} = (P_t - N_p) + H_p \]  

(2.1)

where \( P_t \) is the number of polyps at any time \( t \), \( N_p \) is the nudibranch predation rate in numbers of polyps per day and \( H_p \) is the hydroid growth rate in polyps per day.

Stolon growth was also calculated as

\[ S_{t+1} = (S_t - N_s) + H_s \]  

(2.2)

where \( S_t \) is the volume of stolon at any time \( t \), \( N_s \) is the nudibranch predation rate in mm\(^3\) of stolon per day and \( H_s \) is the hydroid growth rate in volume of stolon per day.
Simulations were started at time zero with 25 polyps or 10 mm$^3$ of stolon tissue and one newly laid nudibranch egg. These values correspond to the mean size of the hydroid colonies used in the predation study. The simulations were run for a total of 28 days which was the maximum time that a nudibranch lived.

**Analysis.**

Statistical analyses were performed using SYSTAT (Systat Inc., Evanston, IL). The relationship between nudibranch size and predation rates was investigated with an analysis of variance model (ANOVA) (Sokal and Rohlf, 1981). Tukey HSD was used to compare nudibranch sizes with different feeding rates.

**Results**

**Hydroid Colony Growth**

*Cordylophora lacustris* grew best at the lower salinities (Figure 2.1). The greatest increase in numbers of polyps occurred at 5 °/oo, with slower growth at 10, 15 and 20 °/oo (Figure 2.1A and B). Some growth occurred at 0 and 25 °/oo. *Cordylophora* colonies did not survive at 30°/oo and shrank in a matter of days.

Percent stolon growth closely followed growth of polyps, with the best growth occurring at 10, 15 and 20 °/oo (Figure 2.1C and D). At these salinities, stolons grew 2-3 mm$^3$/day (Table 2.1). Slower growth occurred at 25 °/oo (stolon growth: 0.50 ± 0.32 mm$^3$/day). Note that stolon growth was not followed at 0, 5, and 30 °/oo.

**Nudibranch Life History.**

The results of the life history study are presented in Table 2.1 and Figure 2.2. The generation time from egg to egg was approximately 17.6 ± 0.3 days (n = 20) and the life cycle from egg to death was 24.5 ± 1.0 days (n = 10). Hatching occurred in 6.3 ± 0.2 days (n = 56). Juveniles metamorphosed in 7.1 ± 0.2 days (n = 45) at a size of 0.3 ± 0.1 mm (n = 20). *Tenellia adspersa* grew exponentially in size until sexual maturity. Size at maturity was 4.5 ± 0.2 mm (n = 20). The growth rate decreased after sexual maturity until a maximum length of 6.1 ± 0.3 (n = 10) was
achieved. Nudibranchs decreased in size near the end of their life, reaching a length of 5.4 ± 0.3 mm (n = 10). This decrease became apparent up to seven days before death.

Mature nudibranchs produced 5.3 ± 0.9 spawn masses per day (n = 10) with 32.1 ± 0.6 eggs per spawn (n = 10). Their lifetime fecundity was 39.4 ± 7.6 spawn per individual (n = 10 for a total of 1408.4 ± 102.9 eggs per individual (n = 10).

**Nudibranch Feeding Rate**

*Tenellia adspersa* feeds by piercing the stolon with its radula and sucking out tissue. Larger nudibranchs (> 6 mm) were observed to feed on the polyps themselves by taking bites out of the polyp. Newly metamorphosed juveniles were invariably found near new hydroid growth. No observable loss of hydroid tissue was observed for the first 4-5 days following metamorphosis. Nudibranchs ate significantly more polyps and more coenosarc as they grew (ANOVA: polyp predation, $F_{4,21} = 168.0$, $p < 0.0001$; stolon predation, $F_{4,24} = 22.9$, $p < 0.0001$) (Figure 2.3). Mature nudibranchs (> 4 mm) consumed 18.0 ± 1.9 mm$^3$ of stolon tissue per day and 6.6 ± 0.2 polyps per day.

**Hydroid Growth Rate**

Hydroid growth rates in terms of polyp production ($H_p$) and stolon production ($H_s$) are presented in Table 2.1. Because nudibranch predation rates increased with increasing size, nudibranch predation rates ($N_p$ and $N_s$) were allowed to increase according to the schedule in Table 2.2. With the exception of the colony at 5% salinity, all colonies were completely eaten within one generation of the nudibranch (Figure 2.4A). These colonies persisted from 10 to 23 days with the colonies at 10-20% salinity persisting longer than ones at higher salinities. Colonies at 5% salinity continued to grow after the end of the time the nudibranch was alive. The pronounced change in polyp production observed in all but the extreme salinities corresponds to
when the nudibranch reached maturity and increased their polyp consumption by 3-fold.

The effect of predation on stolon production closely followed polyp production (Figure 2.4B). Colonies were completely consumed within a single nudibranch generation at all salinities simulated. Colonies persisted from 12 to 18 days with colonies at salinities of 15-20 °/oo persisting longest. The sharp transition from positive growth to negative growth at 10 days is due to the nudibranchs reaching 1 mm in size and increasing their consumption of stolon by 10 fold.

Discussion

The hydroid. *Cordylophora* lives in fresh or brackish water conditions and can tolerate greater fluctuations in its habitat than its marine relatives (Fulton, 1962). In the Great Bay Estuary, *Cordylophora* is only found in low salinity riverine systems and does not occur within the Great Bay (personal observation). The hydroid growth study supports these observations. The best growth rate in terms of polyps occurred at 5°/oo salinity with slower growth occurring at salinities of 10-20°/oo. In terms of change in stolon, good growth occurred at salinities of 10-20°/oo. Growth of stolons was not measured at 5°/oo salinity; however, it is suspected that highest stolon growth would also occur at this salinity. Under controlled conditions, using defined media. Fulton (1960) grew colonies of *Cordylophora lacustris* and found that polyps increased exponentially with a doubling time of 3 days. Stolons grew linearly with a growth rate of 0.1 mm/hr (Fulton, 1963). This translates to 1.5 mm³/day (using a stolon diameter of 0.2 mm (Fulton, 1961)). The present study yielded values higher than Fulton’s (1961) for the hydroid in salinities less than 20°/oo.

In comparison, the nudibranch *Tenellia adspersa* has been found in salinities down to 8°/oo (Table 2.3). However, it appears to be found most frequently at salinities above 15°/oo. Rasmussen (1944) successfully developed eggs from *Tenellia pallida* at a salinity of 20°/oo. Harris et al., (1980) studied the life history of *Tenellia*
_fuscata_ (=*Tenellia adspersa*) under varying temperature and salinity regimes and found that although adult nudibranchs could survive and reproduce at lower salinities (10°/oo), development was incomplete or did not occur. In addition, developing embryos were not able to tolerate changes in salinity until after they began to rotate in their capsules (i.e., after trophophore stage). These observations strongly suggest that *Tenellia* can not survive and produce viable offspring at salinities less than 10-15°/oo.

A variety of mollusks, including the gastropod *Aplysia californica* and the bivalves *Mercenaria mercenaria* and *Bankia gouldi* require a minimum of 4-5 p.p.m. of the element strontium for normal mineralization of shells and statoliths (Gallager, _et al._, 1988). Seawater less than 15°/oo salinity contains less than 5 p.p.m. of strontium (Kuzirian, pers. communication).

The growth dynamics of *Tenellia adspersa* presented in this study are similar to those found by Harris _et al._ (1980) for this species and by Rasmussen (1944) for *T. pallida*. However, the generation time and life cycle were shorter than previously observed. This may be a result of varying laboratory conditions. *Tenellia adspersa* grew exponentially until about the time of the first egg mass. Growth rates decreased until about the 24th day when growth rates were negative. As with previous studies, a great deal of variation in fecundity among individuals was evident. The present study did not compare the life history of *Tenellia* at different salinities. However, Harris _et al._ (1980) did and found very little difference in growth dynamics.

In addition to the hydroids' ability to tolerate lower salinities compared to the nudibranch, *Cordylophora* may benefit from faster growth rates at these lower salinities. A comparison of nudibranch predation rates with hydroid growth rates indicate that once individual nudibranchs grow to 1 mm or greater they can consume more stolon tissue per day than the hydroid growth rate at all salinities measured (10-25°/oo). However, in terms of number of polyps eaten per day, polyp production at lower salinities (5-20°/oo) was faster than the feeding rates of juvenile nudibranchs.
But, once the nudibranch matured, its feeding rate was much higher than hydroid growth rates. At 5°/oo, the rate of polyp production was higher than feeding rate of any sized nudibranch. At higher salinities (25-30°/oo) polyp production was much slower than the feeding rate of any-sized nudibranch. Turpaeva (1963) demonstrated that a single individual of *Tenellia adspersa* could destroy a colony of the hydroid, *Perigonimus megas* consisting of up to 100 polyps in 24 hours (Cited in Roginskaya, 1970).

*Tenellia* produces both pelagic lecithotrophic larvae and capsular metamorphic juveniles, so some of the offspring will remain within the hydroid colony. Coupled with the short generation time observed (2-3 weeks), nudibranchs will build up within a colony very quickly. This will affect predation rates and ultimately the persistence of the hydroid colony.

Another factor affecting the persistence of a hydroid colony is the initial hydroid colony size. The later the initial recruitment of nudibranchs, the larger the hydroid colony will grow and the longer it will persist. This will be particularly important at lower salinities where hydroid growth is faster and where the nudibranch may be stressed.

Both the salinity and predation data have important implications for the distribution and persistence of *Cordylophora* colonies in the Great Bay Estuary. Given the developmental strategy and dispersal characteristics of the nudibranch (discussed in chapter 4) any hydroid colony existing above 10-15°/oo salinity will be quickly colonized by nudibranchs and ultimately will be removed. At higher salinities, colony growth rates are much slower, so persistence of these colonies will be shorter. Colonies below 10-15°/oo salinity may have a refuge by their ability to survive and grow faster than nudibranch predation rates and due to the inability of the nudibranch to tolerate these lower salinities. In addition, the number of nudibranchs in these
colonies will depend solely on the influx of larvae from outside the colony because the nudibranchs within the colony will not be able to reproduce.

When a *Cordylophora* colony is destroyed by *Tenellia*, the perisarc remains. This has implications for the ultimate effect of the nudibranchs on the recruitment of other organisms. Although the three-dimensional structure, and hence effects on current flow over the colony, remains much the same, a healthy hydroid colony, with its polyps intact, will very likely impede or facilitate settling of larvae quite differently than will its skeleton (Standing, 1976)

In summary, the growth and predation patterns documented in this study illustrate both a very dynamic life history for a hydroid and a dynamic and flexible life history for a nudibranch predator specializing on the exploitation of ephemeral prey populations in a constantly changing estuarine environment.
Table 2.1. Growth rate of *Cordylophora lacustris* under varying salinity regimes. Presented as mean (± standard error), n = 10 replicates per treatment. nd = data not collected.

<table>
<thead>
<tr>
<th>Salinity (‰)</th>
<th>(H_p), Change # polyps (polyps * day(^{-1}))</th>
<th>(H_s), Change in Stolon Volume (mm(^3) * day(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.39 (0.13)</td>
<td>nd</td>
</tr>
<tr>
<td>5</td>
<td>8.70 (3.42)</td>
<td>nd</td>
</tr>
<tr>
<td>10</td>
<td>1.58 (0.39)</td>
<td>2.09 (0.53)</td>
</tr>
<tr>
<td>15</td>
<td>1.41 (0.52)</td>
<td>3.75 (1.72)</td>
</tr>
<tr>
<td>20</td>
<td>2.51 (0.74)</td>
<td>3.65 (1.20)</td>
</tr>
<tr>
<td>25</td>
<td>0.18 (0.12)</td>
<td>0.50 (0.32)</td>
</tr>
<tr>
<td>30</td>
<td>-0.20 (0.47)</td>
<td>nd</td>
</tr>
</tbody>
</table>
Table 2.2. Predation rates on *Cordylophora lacustris* by *Tenellia adspersa*. Presented as mean (± standard error) sample size.

<table>
<thead>
<tr>
<th>Size (mm)</th>
<th>Age (days)</th>
<th>Nₚ, polyp predation (#polyps eaten * day⁻¹)</th>
<th>Nₛ, stolon predation (mm³ stolon eaten * day⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-metamorphosis 0-6</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>&lt;1</td>
<td>7-10</td>
<td>0.16 (0.09) 11</td>
<td>0.30 (0.09) 11</td>
</tr>
<tr>
<td>1-2</td>
<td>11-13</td>
<td>0.87 (0.21) 5</td>
<td>4.68 (1.26) 8</td>
</tr>
<tr>
<td>2-3</td>
<td>14</td>
<td>1.55 (0.15) 4</td>
<td>4.89 (1.16) 4</td>
</tr>
<tr>
<td>3-4</td>
<td>15</td>
<td>2.80 (0.25) 3</td>
<td>7.42 (2.23) 3</td>
</tr>
<tr>
<td>&gt;4</td>
<td>16-28</td>
<td>6.60 (0.19) 3</td>
<td>18.01 (1.87) 3</td>
</tr>
</tbody>
</table>

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Table 2.3. Salinity and abundance of *Tenellia* spp. throughout the world.

<table>
<thead>
<tr>
<th><em>Tenellia</em> Species</th>
<th>Locality</th>
<th>Salinity (o/oo)</th>
<th>Availability</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>pallida</em></td>
<td>Georgetown County, SC</td>
<td>20-35</td>
<td>nd</td>
<td>Eyster, 1979</td>
</tr>
<tr>
<td><em>pallida</em></td>
<td>Chesapeake Bay, MD</td>
<td>12.6</td>
<td>nd</td>
<td>Marcus, 1972</td>
</tr>
<tr>
<td><em>pallida</em></td>
<td>Azov Sea, Russia</td>
<td>8</td>
<td>nd</td>
<td>Turpaeva, 1963*</td>
</tr>
<tr>
<td><em>pallida</em></td>
<td>Copenhagen, Denmark</td>
<td>12</td>
<td>nd</td>
<td>Rasmussen, 1944</td>
</tr>
<tr>
<td><em>fuscata</em> (= adspersa)</td>
<td>Isenfjord, Denmark</td>
<td>20</td>
<td>nd</td>
<td>Rasmussen, 1944</td>
</tr>
<tr>
<td><em>fuscata</em></td>
<td>Mystic River, CT</td>
<td>30</td>
<td>year round</td>
<td>Clark, 1975</td>
</tr>
<tr>
<td><em>fuscata</em> (= adspersa)</td>
<td>Thames River, CT</td>
<td>16</td>
<td>nd</td>
<td>Clark, 1975</td>
</tr>
<tr>
<td><em>fuscata</em> (= adspersa)</td>
<td>Great Bay Estuary, NH</td>
<td>20-30</td>
<td>nd</td>
<td>Harris <em>et al.</em>, 1980</td>
</tr>
<tr>
<td><em>adspersa</em></td>
<td>Portsmouth Harbor, NH</td>
<td>30</td>
<td>June to July</td>
<td>Lambert, 1985</td>
</tr>
<tr>
<td><em>adspersa</em></td>
<td>Great Bay estuary, NH</td>
<td>20-30</td>
<td>June to August</td>
<td>Chester, unpublished data</td>
</tr>
</tbody>
</table>

* in Roginskaya (1970). nd = no data.
Figure 2.1. Growth of *Cordylophora lacustris* at a range of salinities, measured as number of polyps (A), change in polyps per day (B), volume of stolon (C) and change in volume of stolon per day.
Figure 2.2. Nudibranch size and fecundity (A) and survivorship data (B) for *Tenellia adspersa* raised on *Cordylophora* at 30\%/oo salinity and 25°C.

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Figure 2.3. Predation rates for *Tenellia adspersa* feeding on *Cordylophora lacustris* in terms of number of polyps eaten per day (A) and volume of stolon eaten per day (B). The results of a Tukey HSD are presented as asterisk if significantly different and lines in not significantly different at $\alpha = 0.05$ level.
Figure 2.4. Simulation comparing the number of polyps (A) and the volume of stolon (B) produced by the hydroid *Cordylophora lacustris* under varying salinity regimes and with the addition of a single nudibranch predator. All simulations began with 25 polyps or 10 mm$^3$ of hydroid stolon tissue and a single nudibranch egg.

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CHAPTER III
LIFE HISTORY OF *Tenellia adspersa* ON DIFFERENT PREY:
GROWTH RATE VERSUS FECUNDITY

**Introduction**

Life history strategies are suites of traits that are co-adapted such that altering one trait may result in concomitant changes in other related traits (Stearns, 1980). A critical event in an organism's life cycle is maturation. Maturation is a costly process that requires resources stored or previously available for somatic growth and maintenance to be diverted to gonad growth and gamete production (Bernardo, 1993). As a result, age and size at maturity can have a large impact on fitness across a wide range of life histories (Stearns, 1992).

Size and age at maturity have been shown to vary among closely related species, among populations within species, and among individuals within populations (e.g., Bell, 1980; Stearns, 1983). Stearns and Koella (1986) distinguished five patterns of age and size at maturity. Organisms can 1) mature earlier at a smaller size, 2) mature at an equal age but a smaller size, 3) mature later at a smaller size, 4) mature later at an equal size, or 5) mature later at a larger size.

Age and size at maturity represents a trade-off between survival and reproduction. Early maturing organisms may benefit by spending a shorter period as a juvenile, resulting in higher survival to maturity (Bell, 1980). They may also benefit because of a shorter generation time (Cole, 1954). Conversely, delaying maturity may allow for further growth and a higher initial fecundity, if fecundity is related to body size. Delaying maturity may also lead to a higher quality of offspring produced, thereby reducing the instantaneous juvenile mortality rate (Stearns, 1980).

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Additionally, delaying maturity may allow an organism to have more reproductive events, as a result lifetime fecundity is increased (Stearns, 1980).

Much of the variability in size and age at maturity can be accounted for by phenotypic plasticity in maturation (Stearns, 1980). Phenotypic plasticity (multiple phenotypes produced by a single genotype) represents the sensitivity of the phenotype to the environment and is determined by the interaction of the genotype with the environment (Caswell, 1983). Because, age and size at maturity are dependent on the individual's growth and mortality rates (Perrin and Rubin, 1990), variation in growth and mortality rates can cause variation in optimal population age and size at maturity. In spatially or temporally changing environments, phenotypic plasticity in maturation may be advantageous by allowing an organism to survive in a wider range of habitats. In these changing environments, natural selection should favor genotypes that present adaptive phenotypic plasticity for these traits, particularly if the environments fluctuate more rapidly than the generation time of the organism (Bernardo, 1993; Perrin and Rubin, 1990).

Tenellia adspersa is a small (5-7 mm) aeolid nudibranch found commonly along the Western Atlantic coast. Tenellia adspersa is a subannual species with a generation time of 20 days from egg to egg when raised at 20°C and 30 °/oo salinity on the hydroid, Cordylophora lacustris (Harris, et al., 1980). At this temperature, development takes place in 7-10 days. Developmental mode is plastic, with a portion of the veligers within a spawn metamorphosing within the capsule (Chapter Four). The rest hatch as non-feeding pelagic larvae that settle and undergo metamorphosis within 24 hours if hydroid is available.

Unlike other aeolids, which feed only on a limited number of hydroids, Tenellia feeds on a wide variety of gymnoblastic and calyptoblastic hydroids (Clark, 1975; Chester, Personal Observations). The majority of these hydroids are seasonally abundant in the Great Bay Estuary, New Hampshire (70°55'N 43°5'W) where their
distribution and abundance can change in as rapidly as one week (Chapter One). In addition, predation by *Tenellia* may eliminate the hydroid within a single generation of the nudibranch (Chapter Two). Therefore, hydroid prey abundance will have an effect on the nudibranchs' life history.

The objective of this study was to determine the effect of hydroid prey species on the nudibranch life history. This was accomplished by culturing *Tenellia adspersa* on three species of hydroids, and comparing life history traits.

**Materials and Methods**

Both the hydroid, *Cordylophora lacustris*, and the nudibranch, *Tenellia adspersa*, were collected and cultured in the lab as in Chapter Two. *Obelia commissuralis* appeared in the aquaria in August 1992. *Hydractinia* sp. was collected in the fall of 1992 on hermit crab shells from Gosport Harbor, Isles of Shoals (New Hampshire) (70°36'N, 43°58'W) and from a large colony existing on the commercial fishing pier in Portsmouth Harbor (New Hampshire) (70°44'N, 42°05'W). All hydroids were cultured using the standard culture methods in Chapter Two, except *Obelia* and *Hydractinia* were maintained at 25°C nat 25°/oo salinity.

Twenty adult nudibranchs were taken from the stock culture, paired and put on slides containing one of the three hydroids at 25°C and 25°/oo. After five days, 20 newly laid spawn masses (2 from each pair) were collected and the number of eggs per mass counted. These were cultured on their respective hydroid and provided with an *ad libitum* amount of hydroid. Water was changed and individuals counted daily. Nudibranch length was measured daily using a Wild dissecting microscope equipped with an ocular micrometer. At one mm, juvenile nudibranchs were separated into pairs. Once maturity was reached, spawn masses were removed weekly and the number of eggs per mass counted. The number of eggs per spawn was not measured for *Tenellia* on *Hydractinia*. Nudibranchs were followed until death.
Statistical analyses were performed using SYSTAT (SYSTAT Inc., Evanston, IL). An analysis of variance model (ANOVA) was used to compare life history traits. Reproductive output is related to size in Tenellia, therefore, an analysis of covariance (ANCOVA) with nudibranch size as a covariate was used to compare reproductive output (number of eggs per spawn and number of eggs) on the two hydroid species (Sokal and Rohlf, 1981).

Life tables and age-classified matrices were constructed to compare population growth on Cordylophora and Obelia (Caswell, 1989; Stearns, 1991). This could not be done for Tenellia on Hydractinia because complete data on reproduction were lacking. Leslie matrices for age intervals of one week were calculated from these life tables following Caswell (1989). Survival probabilities, $P_i$, and age-specific fecundities, $F_i$, were calculated using birth-flow probabilities. Matrices were analyzed using RAMAS/Stage (Applied Biomathematics, Setauket, NY) to obtain population growth rates, stable age distributions, sensitivities and elasticities.

Results

Life Cycle

The life cycle from fertilization to death of Tenellia adspersa was shortest on Cordylophora (Figure 3.1). In all cases, growth was sigmoidal. Hatching and metamorphosis occurred at the same time on Cordylophora and Obelia, but approximately 3 days later on Hydractinia (Figure 3.2A, Table 3.1). Juveniles metamorphosed at the same size on all three hydroid species (0.309 ± 0.049 mm, n = 27) (Figure 3.2B, Table 3.1).

Maturity, as indicated by first spawn, occurred earlier on Cordylophora, compared to nudibranchs on Obelia or Hydractinia (Figure 3.2A, Table 3.1). Nudibranchs matured at a large size on Cordylophora, a medium size on Obelia and at a small size on Hydractinia (Figure 3.2B, Table 3.1). Nudibranchs achieved similar
maximum size on both *Cordylophora* and *Obelia* but never reached as large a size on *Hydractinia*. In all cases, size decreased during the last 3-7 days before death. Size at death did not differ on *Cordylophora* or *Obelia* but was significantly smaller on *Hydractinia*. However, *Tenellia* lived approximately twice as long when raised on *Obelia* and *Hydractinia* as on *Cordylophora*.

**Fecundity**

Lifetime fecundity was lower on *Cordylophora* compared to *Obelia* but the differences were not significant (Figure 3.2C, Table 3.1). Number of eggs were not measured for *Tenellia* on *Hydractinia*. *Tenellia* produced the greatest number of spawn per day on *Cordylophora*, fewer on *Obelia* and fewest on *Hydractinia* (Figure 3.2D, Table 3.1). There was no relationship between the number of spawn produced per day and nudibranch size for *Tenellia* raised on any hydroid species (*Cordylophora*: $r = 0.12$; *Obelia*: $r = 0.43$; *Hydractinia*: $r = 0.15$). *Tenellia* produced fewer spawn per individual on *Cordylophora* and *Hydractinia* than on *Obelia*.

Fecundity in terms of both the number of eggs produced per day and the number of eggs per spawn was positively related to nudibranch size for *Tenellia* on *Cordylophora* and *Obelia* (Figure 3.3A and B). For the relationship between the number of eggs produced per day and nudibranch size, the correlation coefficient ($r$) for *Tenellia* raised on *Cordylophora* was 0.61 and 0.75 on *Obelia*, indicating a good fit to the data (Figure 3.3A). The slopes of these two regressions were homogeneous (ANCOVA: $F_{1,166} = 2.13$, $p = 0.146$). With the effect of nudibranch size removed, *T. adspersa* produced more eggs per day when raised on *Cordylophora* compared to *Obelia* (ANCOVA: $F_{1,167} = 139.20$, $p < 0.0001$). Correlation coefficients ($r$) for the relationship between the number of eggs per spawn and nudibranch size were slightly higher ($r = 0.88$ for *Tenellia* raised on *Cordylophora* and $r = 0.91$ for *Tenellia* raised on *Obelia*), and also indicated a good fit to the data (Figure 3.3B). The slopes were
homogeneous (ANCOVA: $F_{1.166} = 1.0, p = 0.32$). *Tenellia* produced more eggs per spawn on *Cordylophora* than on *Obelia* (ANCOVA: $F_{1.167} = 221.5, p < 0.0001$).

**Demographic Analysis.**

Survivorship with respect to age was calculated for nudibranchs raised on *Cordylophora, Obelia* and *Hydractinia*. Nudibranchs living on *Obelia* experienced higher survival than ones living on *Cordylophora* or *Hydractinia* (Figure 3.4A). Survival of the younger age classes declined sharply for nudibranchs raised on *Hydractinia* compared to nudibranchs raised on the other two hydroids. Hatching and metamorphosis occurred between seven to twelve days on all hydroids. This period, between hatching and metamorphosis appeared to be the critical survival period for *Tenellia* on all three hydroids. In addition, survival declined with age of nudibranchs. However, the decline was sharper for *Tenellia* on *Hydractinia* and *Cordylophora* compared to *Tenellia* on *Obelia*.

A demographic analysis was only possible for *Tenellia* on *Cordylophora* and *Obelia* because of the lack of a complete data set for *Tenellia* on *Hydractinia*. Age-specific fecundity ($M_x$) schedules for *Tenellia* on the two hydroid prey revealed earlier and higher reproductive output for *Tenellia* on *Cordylophora* (Figure 3.4B). However, the duration was longer for *Tenellia* on *Obelia*.

Finite population growth rates for the two populations of nudibranchs were similar but higher for *Tenellia* on *Obelia* (Table 3.2). Mean generation times for nudibranchs raised on *Cordylophora* was 2 weeks less than for nudibranchs raised on *Obelia*. Net reproductive rates were higher for nudibranchs on *Obelia* compared to nudibranchs on *Cordylophora*.

Predicted stable age distributions for *Tenellia* on the two hydroid prey indicated that both populations were dominated by embryos and larvae, with less than 1% of the population above the mean reproductive age (Figure 3.5A).
Patterns of change in age-specific reproductive values were higher and peaked earlier for *Tenellia* raised on *Cordylophora* (Figure 3.5B). Peak reproductive values were reached earlier when *Tenellia* was raised on *Cordylophora* (3 weeks) than for *Tenellia* on *Obelia* (5 weeks).

Fertility elasticities ($F_i$) were lower than survival elasticities ($P_i$) for *Tenellia* raised on both species of hydroid (Figure 3.6). On both hydroids, the population growth rates ($\lambda$) were more sensitive to changes in early survivorship ($P_i$). In addition, changes in the earliest age classes (one week or less) were more important for *Tenellia* on *Cordylophora* than on *Obelia*. Population growth rates were also more sensitive to changes in fecundity for *Tenellia* on *Cordylophora* compared to nudibranchs on *Obelia*.

**Discussion**

In fluctuating environments, phenotypic plasticity in age and size at maturity can evolve if the environments fluctuate more rapidly than the generation time of the organism. *Tenellia adspersa* preys on a variety of gymnoblastic and calyptoblastic hydroids (Clark, 1975). The majority of these hydroids are patchily distributed and temporally unpredictable. Their abundance and distribution within the Great Bay Estuary can change in as little as one week (Chapter One), which, as demonstrated here (see also Harris, *et al.*, 1980) is within the generation time of *Tenellia*. This study also demonstrated that size and age at maturity in *Tenellia* varied with hydroid prey. Stehr (1964) hypothesized that if the periodicity of environmental change is such that it falls within the generation of an organism living in that environment then selection will favor mechanisms for maintaining variation in the organism's life history traits. *Tenellia* might be responding to variation in hydroid diversity and abundance by selecting for a variable life history. Roff (1978) predicted that spatio-temporal heterogeneity in environmental conditions can influence fluctuations in body size if the environmental heterogeneity gives rise to variable size dependent mortality rates.
Survivorship values for *Tenellia* on both *Cordylophora* and *Obelia* declined with age and size. However, survivorship declined more slowly on *Obelia* compared to nudibranchs raised on *Cordylophora*.

The results presented here indicate that projected populations of *Tenellia* grew at similar rates when living on *Cordylophora* as when living on *Obelia*. Survival was lower but fecundity higher when *Tenellia* was raised on *Cordylophora*. In contrast, *Tenellia* experienced higher survival but lower fecundity on *Obelia*. In addition, lifespan was approximately twice as long for obeliid nudibranchs compared to ones living on *Cordylophora*. This suggests a trade-off between adult survival and reproduction. Assuming that an organism has a finite amount of resources available for maintenance and reproduction then, under resource limited conditions, an increased allocation to reproduction will result in a decreased allocation to maintenance and as a consequence, reduced survival. Snell and King (1977) found an inverse relationship between reproduction and lifespan in the rotifer, *Asplanchna brightwellii*. Haukioja and Hakala (1978) found an inverse relationship between reproductive effort and length of reproductive lifespan in comparisons of 13 populations of the bivalve, *Anodonta piscinalis*.

This study demonstrated a relationship between fecundity and size. Larger nudibranchs produced more eggs per spawn and more eggs per day than smaller nudibranchs. This relationship has been demonstrated in other gastropods (e.g., Spight and Emlen, 1976), in most invertebrates (e.g., Fritz and Morris, 1985; Gliwicz and Lampert, 1994; Kessler, 1971; Peterson, 1950) and in vertebrates (Stearns, 1992).

Nudibranchs living on *Obelia* and *Hydractinia* matured later and at a smaller size than ones living on *Cordylophora*. Berrigan and Charnov (1994) suggested that decreased food quality reduces growth rate and typically results in delayed maturation at a smaller size. Qian and Chia (1992) found that the polychaete, *Capitella* when fed...
four diets differing in nutritional quality showed differences in growth, maturity, fecundity, egg size and egg organic content that were related to the caloric content of the diet. Nudibranchs raised on Obelia and Hydractinia grew in size at a slower rate compared to nudibranchs raised on Cordylophora. Nakaoka and Matui (1994) found that the growth rate of the bivalve Yoldia notabilis was positively correlated with chlorophyll a content, suggesting that growth rate was dependent on variation in available food. In the wolf spider Gemlycosa domiflex, the amount of available food affected both egg production and body size (McQueen, 1983). The three hydroids used in this experiment differ in their morphology. Both Cordylophora and Obelia produce arborescent colonies with a number of upright branches terminating in polyps arising from stolons running along the substratum. The stolons and uprights are surrounded by a non-living chitinous envelope called the perisarc. In contrast, Hydractinia produces colonies with closely placed naked polyps arising from a mat of stolons that are fused together forming a hydrorhizal plate. In Obelia, the polyps are small and surrounded by a cup-shaped theca whereas in Cordylophora and Hydractinia the polyps are larger and athecate. Additionally, the diameter of the stolons are smaller in Obelia compared to Cordylophora (Cordylophora, 294.1 ± 7.6 μm, n=60; Obelia, 266.7 ± 9.1 μm, n=49. Comparison, t=2.33, d.f=107, p<0.05). Differences in nutritional quality between these three hydroid prey may exist as well.

Reduced feeding rates may also reduce nudibranch growth rates with the same result of delayed maturation at a smaller size. Tenellia differed in its feeding behavior on the three hydroids. When feeding on Cordylophora, nudibranchs pierced the perisarc and sucked out coenosarc tissue (Chapter Two). Larger individuals were also observed to feed directly on polyps. When feeding on Obelia, nudibranchs were only observed piercing the perisarc and suck tissue from the stolons and uprights. When feeding on Hydractinia, nudibranchs would rasp the naked tissue from the edge of a growing colony. The smaller diameter of the perisarc would have made it more
difficult for *Tenellia* to remove coenosarc from the stolon and uprights of *Obelia* and, thus, reducing feeding rates. Nudibranchs were rarely seen crawling within *Hydractinia* colonies, and all tissue loss occurred on the edges of the colonies, suggesting that feeding rates on *Hydractinia* may have been lower as well.

Growth rates of 15-20 mm/day were measured by Berrill (1949) for *Obelia commissularis* at 20-25°C and 25-30°/⁰⁰ salinity. These values can be converted to 0.83-1.11 mm³/day of stolon volume using the above diameter. The resulting value was much greater than the one for *Cordylophora* raised under the same conditions (*Cordylophora* growth rate = 0.50 mm³ stolon/day, Chapter Two). It was suggested in the preceding chapter (Chapter Two) that *Tenellia* could remove a *Cordylophora* colony in a short period of time, depending on initial hydroid colony size and number of nudibranchs within a colony. Given the faster growth of *Obelia*, these colonies should persist longer before being removed by predation.

Although, colonies of all three hydroids are be found in the same type of habitats (piers, floats and other artificial and natural structures), *Hydractinia* also occurs on hermit crab shells. In addition, the three hydroids differ in their distributions. *Cordylophora* is found in freshwater to low salinity riverine systems (Chapter Two). *Obelia* is found in moderate to high salinities found in the Great Bay Estuary and out on the open coast. *Hydractinia* is only found on the open coast at higher salinities.

Environmental stability also varies with location within the estuary. The riverine systems where *Cordylophora* is predominantly found are highly variable and can drastically change in a short period due to run-off from storms. The higher salinities where *Obelia* is commonly found are less variable. Salinity in open coastal systems where *Hydractinia* occurs is the least variable. Although *Tenellia* can survive at salinities below 10-15°/⁰⁰, it cannot successfully reproduce at salinities below 15°/⁰⁰ (Harris et al., 1980). Though *Cordylophora* may be a higher quality food source than
Obelia or Hydractinia its collection by the nudibranch may involve additional risks, some of them due to environmental instability. A life history strategy incorporating a shorter generation time would be advantageous in minimizing that risk. An additional instability or risk factor is due to the ephemeral nature of the hydroids. Thus, the flexible life history would also be advantageous for survival in such environments.

In conclusion, this study demonstrates that Tenellia adspersa has adopted a very flexible and dynamic life history for survival within the estuary where environmental conditions are variable and continually changing.
Table 3.1. Summary of life history data for *Tenellia adspersa* on *Cordylophora lacustris*, *Obelia commiseralis* and *Hydractinia* sp.. Data from nudibranchs cultured at 25°C and 25°/oo salinity and provided with an *ad libitum* amount of hydroid.

<table>
<thead>
<tr>
<th></th>
<th><em>Cordylophora lacustris</em></th>
<th></th>
<th><em>Obelia commiseralis</em></th>
<th></th>
<th><em>Hydractinia sp.</em></th>
<th></th>
<th>ANOVA Results</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean s.e. n</td>
<td>Mean s.e. n</td>
<td>Mean s.e. n</td>
<td>F-ratio d.f P</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Time (days) to:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hatching</td>
<td>6.04 0.22 12</td>
<td>5.48 0.24 10</td>
<td>10.17 0.54 6</td>
<td>25.64 2,22 ***</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Metamorphosis</td>
<td>6.92 0.22 12</td>
<td>6.41 0.24 10</td>
<td>11.67 0.49 6</td>
<td>21.20 2,22 ***</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maturity</td>
<td>17.57 0.62 10</td>
<td>25.31 0.69 8</td>
<td>29.67 0.92 6</td>
<td>36.60 2,24 ***</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Death</td>
<td>23.86 1.48 5</td>
<td>51.61 1.66 4</td>
<td>52.17 4.96 6</td>
<td>8.90 2,19 *</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Length (mm) at:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Juvenile</td>
<td>0.28 0.01 10</td>
<td>0.27 0.01 6</td>
<td>0.35 0.04 6</td>
<td>2.31 2,24 ns</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maturity</td>
<td>4.45 0.19 10</td>
<td>3.53 0.21 8</td>
<td>2.32 0.12 6</td>
<td>25.68 2,24 ***</td>
<td></td>
<td></td>
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<tr>
<td>Maximum Length</td>
<td>5.83 0.30 5</td>
<td>6.25 0.34 4</td>
<td>3.72 0.29 6</td>
<td>11.83 2,19 ***</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Death</td>
<td>5.25 0.44 5</td>
<td>5.25 0.49 4</td>
<td>2.99 0.35 6</td>
<td>12.29 2,19 **</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Fecundity:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td># eggs * Individual⁻¹</td>
<td>1301.60 475.39 5</td>
<td>2543.00 531.50 4</td>
<td>nd</td>
<td>0.01 1,14 ns²</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td># spawn * Individual⁻¹</td>
<td>36.20 15.25 5</td>
<td>97.25 17.05 4</td>
<td>45.00 13.04 6</td>
<td>0.17 2,19 ns</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td># spawn * Day⁻¹</td>
<td>4.67 0.50 5</td>
<td>3.89 0.56 4</td>
<td>1.96 0.29 6</td>
<td>4.44 2,19 *</td>
<td></td>
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<td></td>
</tr>
</tbody>
</table>

ANOVA results (at α = 0.05 level): ns = not significant, * = p < 0.05, ** = p < 0.001, *** = p < 0.0001, nd = no data. *² = comparison using t-test (at α = 0.05 level).
Table 3.2. Summary of demographic data for a number of invertebrate species, including *Tenellia adspersa*. Values for *Tenellia* calculated for 1-wk age intervals.

<table>
<thead>
<tr>
<th>Species</th>
<th>Net Repro. Rate, R₀</th>
<th>Generation Time, T</th>
<th>r</th>
<th>Lifespan</th>
<th>Reference</th>
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</thead>
<tbody>
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<td><strong>Cnidaria</strong></td>
<td></td>
<td></td>
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<tr>
<td><em>Balanophyllia elegans</em></td>
<td>2.7</td>
<td>3.6 years</td>
<td>0.29</td>
<td>6.5-11 yrs</td>
<td>Fadlallah, 1983</td>
</tr>
<tr>
<td><strong>Annelida</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Streblospio benedicti</em> (p)</td>
<td>17.6</td>
<td>15.4 weeks</td>
<td>0.19</td>
<td>36 wks</td>
<td>Levins et al., 1987</td>
</tr>
<tr>
<td><em>Streblospio benedicti</em> (I)</td>
<td>93.4</td>
<td>16.4 weeks</td>
<td>0.28</td>
<td>36 wks</td>
<td>Levins et al., 1987</td>
</tr>
<tr>
<td><em>Capitella capitata</em></td>
<td>36.7</td>
<td>13.8 weeks</td>
<td>0.26</td>
<td></td>
<td>Redman, 1984*</td>
</tr>
<tr>
<td><em>Polydora ligni</em></td>
<td>9.5</td>
<td>8.2 weeks</td>
<td>0.28</td>
<td></td>
<td>Zajak, 1985*</td>
</tr>
<tr>
<td><em>Ophyotrocha</em> sp.</td>
<td>250.8</td>
<td>6.2 weeks</td>
<td>0.89</td>
<td></td>
<td>Akesson, 1982*</td>
</tr>
<tr>
<td><strong>Mollusca</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Helicella pappi</em></td>
<td>3.0</td>
<td>25.7 months</td>
<td>0.04</td>
<td></td>
<td>Lazaridou-Dimitriadou, 1995</td>
</tr>
<tr>
<td><em>Gemma gemma</em></td>
<td></td>
<td></td>
<td>0.18</td>
<td></td>
<td>Weinburg et al., 1986</td>
</tr>
<tr>
<td><em>Doridella steinburgae</em></td>
<td></td>
<td></td>
<td>0.18</td>
<td></td>
<td>Yoshioka, 1986</td>
</tr>
<tr>
<td><em>Corambe pacifica</em></td>
<td></td>
<td></td>
<td>0.16</td>
<td></td>
<td>Yoshioka, 1986</td>
</tr>
<tr>
<td><em>Tenellia adspersa</em> (c)</td>
<td>4.3</td>
<td>2.8 weeks</td>
<td>0.16</td>
<td>4-5 wks</td>
<td>Present study</td>
</tr>
<tr>
<td><em>Tenellia adspersa</em> (o)</td>
<td>5.6</td>
<td>4.9 weeks</td>
<td>0.27</td>
<td>7-8 wks</td>
<td>Present study</td>
</tr>
</tbody>
</table>

(p) = planktotrophic, (I) = lecithotrophic, (c) on *Cordylophora*, (o) = on *Obelia*. * = cited in Levins et al. (1987).
Figure 3.1. Nudibranch size versus age for *Tenellia adspersa* raised on *Cordylophora lacustris*, *Obelia commiseralis* and *Hydractinia* sp.. Bars = standard error of measure.
Figure 3.2. The life history of *Tenellia adspersa* raised on three hydroid prey species. Age (A) and size (B) at metamorphosis, maturity and death, lifetime fecundity (C) and number of spawn per day and per individual (D). Results of ANOVA and Tukey HSD comparisons are presented as a line connecting values not different and an asterisk for values significantly different at $\alpha = 0.05$ level.

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Figure 3.3. Number of eggs per day (A) and number of eggs per spawn (B) versus nudibranch size for *Tenellia adspersa* feeding on two hydroid prey, *Cordylophora lacustris* (solid circles) and *Obelia commiseralis* (open squares).
Figure 3.4. Survivorship (A) and age-specific fecundity (B) for *Tenellia adspersa* feeding on 3 hydroid prey, *Cordylophora lacustris, Obelia commiseralis,* and *Hydractinia* sp.
Figure 3.5. Percent of the population (A) and reproductive value (B) for *Tenellia adspersa* feeding on two hydroid prey, *Cordylophora lacustris* and *Obelia commiseralis*. 

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Figure 3.6. Elasticities of lambda with respect to $P_i$ as a function of age (A) and with respect to $F_i$ as a function of age (B).
CHAPTER IV
DEVELOPMENTAL PLASTICITY IN Tenellia: THE INFLUENCE OF ADULT NUTRITION ON REPRODUCTION AND DEVELOPMENT

Introduction

In many marine invertebrates, egg size is broadly correlated with developmental time, developmental mode (Thorson, 1946; 1950; Rice, 1975; Chia, 1976; Todd, 1981; Hadfield and Switzer-Dunlap, 1984; Emlet et al., 1987) and organic content (Strathmann and Vedder, 1977). However, a large amount of variation in egg size and organic content has been observed within a species and even within a single spawn (Bayne et al., 1978; Turner and Lawrence, 1979; Sinervo et al., 1984; Mcclintock and Pearse, 1986; McEdward and Carson, 1987; Lambert and Todd, 1994). Several studies have demonstrated that the nutritional state of the adults can affect egg size and organic content (Bayne et al., 1975, 1978; Mcillup and Butler, 1979; Thompson, 1983; George, 1994). Variation in maternal investment can impart differences in larval survivorship, size at settlement and other life history characteristics, as demonstrated in prosobranch gastropods (Hughes and Roberts, 1980) and in echinoderms (Sinervo et al., 1984; Sinervo and McEdward, 1988; George, 1994).

Tenellia adspersa is a small (5-7 mm) aeolid nudibranch common in New England estuarine environments (Clark, 1975). It has a subannual life cycle, with a short generation time (20 days from egg to egg when raised on the hydroid, Cordylophora lacustris at 30 °/oo salinity and 20° C) and produces 3-5 spawn per day with 25-50 eggs per spawn (Harris et al., 1980). Tenellia adspersa is a generalist, feeding on a variety of gymnoblastic and calyptoblastic hydroids. The majority of these
Hydroids are seasonally abundant along the New England coasts and estuaries, existing as patches on piers, floating docks, eel grass blades and other natural and artificial structures. Within the Great Bay Estuary (New Hampshire), the distribution and abundance of these hydroids can change within as little as one week (Chapter One), raising the possibility that *Tenellia* could exhaust its food supply within one generation. Developmental mode has been considered to be capsular metamorphic (i.e., metamorphosis within the egg capsule and hatching as a juvenile) (Roginskaya, 1970). However, a switch to pelagic lecithotrophic development has been observed in individuals that had been starved for up to 7 days (personal observation). Developmental plasticity has also been observed in other species of *Tenellia* (Rassmussen, 1944; Eyster, 1979).

The objectives of this study were to determine if food ration affected adult reproduction, if egg size in *Tenellia* was phenotypically plastic and affected by the adult nutritional regime, and if variation in egg size affected development through metamorphosis. These objectives were investigated by comparing adult size, reproductive output, egg size, egg weight, development time and developmental mode in *Tenellia* under different nutritional regimes.

**Materials and Methods**

*Tenellia adspersa* was collected in October 1991 from a floating dock on the Squamscott River, Rockingham, New Hampshire (70° 56' N, 43° 02' W). Stock cultures were established in finger bowls at 25°C and a salinity of 20°/oo. They were fed colonies of the hydroid, *Cordylophora lacustris* as needed. *Cordylophora lacustris* was collected in September 1991 from a floating dock on the Oyster River, Durham, New Hampshire (70° 55' N, 43° 08' W) and was cultured on glass slides that were suspended in aquaria at 25°C and a salinity of 20°/oo. They were fed nauplii of *Artemia salina* daily.
Two experiments were performed to determine the effect of adult nutritional regime on nudibranch reproduction and offspring development; the first investigated variation in developmental time and mode among spawn, and the second investigated variation within spawn. The adult nudibranchs used in both experiments were f₁ progeny taken from five pairs isolated from the stock culture. The f₁ progeny were followed throughout their life cycle to determine the effect of starvation on reproductive output and adult longevity. Adult size, number of spawn masses and number of eggs per spawn were recorded daily. Pairs of nudibranchs were maintained in separate stacking dishes with daily water changes until maturity, when each pair was separated and maintained in an individual dish. Individuals were paired with their mate for four hours each day to allow copulation.

Experiment One.

This experiment was performed in the lab at 25°C and 20⁰/oo salinity. One group of nudibranchs (control group) was provided an ad libitum amount of Cordylophora. A second group (experimental group) was also provided with an ad libitum amount of Cordylophora. However, these nudibranchs were starved for 4 days beginning 3 days after the onset of maturity. Four days was considered a reasonable period for starvation because preliminary observations indicated that changes in offspring development occurred rapidly (within a day) and by the end of starvation, reproductive output had ceased in all experimental nudibranchs.

Each treatment group consisted of twelve randomly selected nudibranchs from the f₁ progeny. Observations began on day 20 (i.e., 20 days since eggs giving rise to the f₁ generation were laid) with starvation beginning on day 22 after that day’s spawn masses were collected. Hydroids were returned to the experimental nudibranchs on day 26. Spawn masses from both groups (control and experimental) were collected daily for a total of 9 days (3 days prior, 4 days during and 2 days after starvation).
masses were observed for an additional two days to determine if reproductive output had recovered. Each spawn mass was moved to a separate container where the number of eggs per spawn was counted at 60x using a Wild dissecting microscope. All spawn masses were videotaped at 40x magnification through a Nikon compound microscope, and videotaped images were transferred to computer using NIH Image, vers 1.57 to measure the maximum diameters of all uncleaved eggs.

Each day, one spawn mass from each individual was used for weight determination. Each spawn mass was rinsed in 3% ammonium formate to remove salts, blotted dry, and dried at 80°C for 48 hours to constant weight. Mean investment per egg was calculated by dividing the total spawn dry weight by the number of eggs and represented the total parental contribution in terms of egg, capsule and egg matrix for each egg. However, this calculation assumes a constant proportion between egg and non-egg components that may not be true (see Discussion).

The remaining (2-3) spawn collected each day were cultured to determine time to hatching, time to metamorphosis and developmental type. Development was considered capsular metamorphic if metamorphosis took place within the egg capsule. A small piece of *Cordylophora* was placed in the culture vessel to promote larval settlement and metamorphosis.

**Experiment Two.**

This experiment followed the same general protocol as the first experiment, with a control group in which nudibranchs were provided with an *ad libitum* amount of hydroid and an experimental group in which nudibranchs were starved beginning 3 days after the onset of maturity. However, this experiment was performed in an environmental chamber at 20°C and a salinity of 20%o, and each treatment consisted of six randomly drawn f1 progeny nudibranchs. Additionally, the experimental group was starved for 7 days at which time all experimental nudibranchs had ceased egg
laying. Spawn masses from both groups were collected for a total of 13 days (3 days prior, 7 days during and 3 days after starvation). Each day, one spawn mass from each individual was removed to separate containers where the number of eggs per spawn were counted and egg diameters measured as in experiment one. All spawn masses were videotaped daily to follow the fate of individual eggs. The resulting number of spawn cultured and videotaped on a daily basis precluded the use of more than one spawn per individual per day.

Data Analysis.

Statistical analyses were accomplished using SYSTAT (SYSTAT Inc., Evanston, IL). Adult life history traits (i.e., age and size at metamorphosis, maturity and longevity) were compared between treatments with t-tests. A two-factor Analysis of Variance (ANOVA) model with treatment and day as factors was used to compare differences in number of spawn masses, number of eggs per mass, developmental times (i.e., times to hatching, to metamorphosis and in plankton), mean egg diameter and mean investment per egg between treatments over the experimental period (Sokal and Rohlf, 1981). Tukey HSD test was used to determine statistical differences between treatment and day. This model allowed comparisons between treatments, prior to, during and after adult starvation. Spawn masses produced by the same individual on subsequent days were assumed independent of each other. An arcsine transformation was used to normalize percentage data (i.e., % developing, % hatching, and % metamorphosing) (Krebs, 1989).

To determine the amount of variability in egg diameter at each level (i.e., within spawn, among spawn within individual, and among individuals), a two-level nested ANOVA with unequal sample sizes was used to compare the diameters of eggs produced by fed adults. The data were taken from a preliminary experiment, under the same conditions as the first experiment (i.e., adults kept at 25°C and 20°/oo with daily
water changes and provided with an *ad libitum* amount of hydroid). The diameters of 6 to 17 eggs from each of three spawn masses were collected from each of 4 nudibranchs.

A two-level nested ANOVA was also used to compare the mean investment of eggs at each level (i.e., within spawn, among spawn within individual, and among individuals). The data were taken from a preliminary experiment in which the adults were cultured under the same conditions as the first experiment. Four spawn from 3 individuals were collected on 3 consecutive days.

**Results**

**Experiment One.**

**Adult Life Cycle.** Growth of the adults was sigmoidal (Figure 4.1). Table 4.1 presents comparisons of control versus experimental nudibranchs for life history traits. Metamorphosis occurred in both treatments around day 8. First spawn mass was produced in both groups around day 19 at 3.60 ± 0.27 mm (n = 20) in length. Experimental nudibranchs decreased in size during starvation and were significantly smaller than control nudibranchs by the end of starvation (day 26). Growth of the experimental nudibranchs resumed one day after food was returned. There was no significant difference in average life span between treatments.

**Fecundity.** Reproductive output (number of eggs produced per day and the average number of eggs per spawn) was positively correlated with adult size (Figure 4.2A and B). However, there was no relationship between adult size and number of spawn produced (r = 0.389).

The effect of starvation on fecundity was examined by comparing the number of spawn masses produced and the average number of eggs per spawn. Control nudibranchs produced significantly more spawn per day than experimental nudibranchs (Figure 4.3A, Table 4.2). Control nudibranchs did not significantly vary in spawn mass production over time, but spawn production tended to increase with age. Spawn
masses produced by the experimental nudibranchs declined sharply from five spawn on day 23 to one spawn on day 24 and continued at this level until day 30, three days after food was returned.

The number of eggs per spawn produced by the control nudibranchs increased over time, with significantly fewer eggs per spawn produced on days 20-21 than on days 23, 29-30 (Figure 4.3B; Table 4.2). The number of eggs per spawn produced by the experimental nudibranchs decreased beginning on day 24 (one day after the onset of starvation) and continuing until 3 days after the end of starvation (day 29). On day 29, experimental nudibranchs produced significantly fewer eggs per spawn than control nudibranchs.

**Egg Diameter.** The nested ANOVA did not detect differences in egg diameters among individuals (Nested ANOVA: $F_{3,137} = 0.648$, $p=0.586$), nor among spawn masses within individuals (Nested ANOVA: $F_{8,137} = 1.513$, $p=0.158$). However, a comparison of the mean-squares revealed that egg diameters were more variable among spawn masses within individuals than among individuals.

Because egg diameter did not significantly vary among either individual or spawn, mean egg diameter was used in the two-factor ANOVA. Experimental nudibranchs produced smaller eggs than control individuals only during starvation (Figure 4.4A, Table 4.2). Mean egg diameters produced by control nudibranchs did not vary over time, except for significant differences between days with the largest diameters (day 21 and 22) and the day with the smallest diameter (day 27). Egg diameters also did not vary between control and experimental nudibranchs either before (day 20, 21 and 22) or after (day 27 and 28) starvation.

**Investment per Egg.** A regression of spawn dry weight versus number of eggs yielded a significant positive relationship with $r = 0.98$. This indicated that little if any inorganic material remained after the ammonium formate rinse. A nested ANOVA of mean investment demonstrated no significant differences among individuals (Nested

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ANOVA: $F_{2,27} = 0.470, p=0.630$). Additionally, no significant differences were observed among days within individuals (Nested ANOVA: $F_{6,27} = 0.620, p=0.713$).

Mean investment per egg followed a similar pattern to mean diameters, but was less variable (Figure 4.4B, Table 4.2). Egg investment by control nudibranchs did not significantly vary over the experimental period and averaged $0.67 \pm 0.02 \mu g$ (note that weights were not measured on day 20 due to a technical error). There was no significant difference in weight between spawn produced by control and experimental nudibranchs during the 2 days prior to starvation (day 21 and 22) and the 2 days after the end of starvation (day 27 and 28). However, during starvation (day 23 to 26) the weight of spawn produced by experimental nudibranchs decreased compared to control nudibranchs.

**Offspring Development.** Development from fertilization to metamorphosis was similar between controls and experimental before and after starvation (Figure 4.5A, Table 4.2). Metamorphosis occurred in 7 days. However, during starvation total development time decreased by approximately 2 days in offspring produced by experimental nudibranchs, and was significantly less than controls on day 25 and 26.

The capsular period of offspring produced by the experimental nudibranchs during starvation was also lower compared to those produced by the control nudibranchs (Figure 4.5B, Table 4.2), and was significantly less than controls on day 24-27. Offspring produced by control nudibranchs did not vary over time, and averaged 6 days to hatching. Also, the capsular period of offspring produced by experimental nudibranchs was not significantly different before starvation (day 20 to 22) or after starvation (day 27 and 28) when compared to those produced by control nudibranchs.

There was no difference in the pelagic period of hatched veligers produced by control nudibranchs during the experimental period, and no significant difference was observed between control and experimental nudibranchs either before (day 20-22) or
after starvation (day 27-28) (Figure 4.5C, Table 4.2). However, pelagic period of veligers produced by the experimental nudibranchs significantly increased during the starvation period when compared to those produced by control nudibranchs.

Twenty to fifty percent of the spawn masses laid by the control nudibranchs contained embryos undergoing capsular metamorphic development. A similar trend was observed in spawn laid by the experimental nudibranchs under ad libitum food (pre and post starvation). Yet, all of the embryos in spawn laid by the experimental nudibranchs under starvation conditions underwent pelagic lecithotrophic development. Survival to juvenile stage was not different in spawn masses produced by fed and starved adults (fed adults = 79%, Starved adults = 90%, Chi-square = 2.12, 1 d.f., p=0.1454).

Experiment Two.

Adult Life Cycle. Adults followed the same pattern as in experiment one but growth was slower (Table 4.1). Metamorphosis in both treatments occurred around day 11. First spawn occurred around day 29, 10 days later than in experiment one. Experimental nudibranchs decreased in size during starvation, and were significantly smaller than controls by the end of starvation. Detectable growth was observed one day after food was returned. Experimental nudibranchs lived nearly a week longer than controls.

Fecundity. A positive relationship was observed between reproductive output (number of eggs and number of eggs per spawn) and nudibranchs size. Correlation coefficients were higher than in the previous experiment (Number of eggs: r = 0.703; Number of eggs per spawn: r = 0.711). No relationship was observed between nudibranch size and the number of spawn masses produced per day (r= 0.226).

Control nudibranchs produced more spawn masses than experimental nudibranchs (Table 4.2). Control nudibranchs did not vary in spawn production over time, and averaged 3-4 spawn per day. Spawn production by experimental nudibranchs
were not different from controls before starvation. During starvation spawn production decreased from 5 to < 1 on the last day of starvation. Number of spawn produced by the experimental nudibranchs increased after food was returned but did not reach 3-4 spawn per day until 4 days after starvation ceased.

Number of eggs per spawn produced by control nudibranchs increased over time, ranging from 31 ± 8 eggs to 64 ± 6 eggs (Table 4.2). Prior to starvation, experimental nudibranchs were not different from controls. During starvation there was a sharp decline in number of eggs per spawn from 46±1 eggs to 1 ± 1 eggs. The number of eggs per spawn increased again after starvation, and five days after food was returned reached 27 ± 4 eggs.

Egg Diameter. Mean diameters of eggs produced by control nudibranchs did not vary over time (126.6 ± 0.6 μm). As in the first experiment, eggs produced by starved adults were significantly smaller (111.9 ± 1.1 μm) than those produced by fed adults (Figure 4.6A, Table 4.2). Eggs produced by control and experimental nudibranchs did not vary before (days 30, 31, and 32) or after (days 40, 41, and 42) starvation.

Offspring Survival. The percent of offspring developing within a spawn did not vary between treatments (Table 4.2). However, there was a general increase in both treatments from 84% on day 30 to 98% on day 42. There was also no interaction between treatment and day.

The percent of offspring within a spawn that survived to hatching did not vary between treatment and day (Table 4.2) and averaged 44% for control and 57% for experimental nudibranchs. There was no interaction between treatment and day.

The percent of offspring reaching metamorphosis were consistently higher for experimental (34%) than for control nudibranchs (24%) (Table 4.2). There were also significant differences in percent metamorphosis with respect to day, with the lowest
(13%) on day 42 and the highest percent (49%) on day 37. However, no interaction between treatment and day was observed.

**Offspring Development.** Development was slower in this experiment compared to experiment one. Total development time was significantly less for offspring produced by experimental nudibranchs during starvation (Table 4.2). Offspring produced by control nudibranchs did not vary and took 10-11 days to reach metamorphosis. Offspring produced by experimental nudibranchs also were not significantly different before or after starvation. However, by the end of starvation, offspring produced by experimental nudibranchs took 8-9 days to reach metamorphosis.

Development to hatching (capsular period) was also longer compared to the previous experiment. Offspring produced by experimental nudibranchs reached hatching sooner than offspring produced by control nudibranchs (Control: 9-10 days. Experimental: 8-9 days) (Table 4.2). Controls were not significantly different over time and were not different from experimental either before or after starvation.

Pelagic period of larvae produced by control nudibranchs did not vary over time, and ranged 1-2 days. Experimental were not different from controls either before or after starvation. However, during starvation, pelagic period increased to 3-4 days and was significantly higher than controls on days 35-36.

The percentage of offspring within a spawn that developed as capsular metamorphic juveniles varied between treatment and day (Figure 4.6B, Table 4.2). Controls did not significantly vary over time, ranging from 7-13% capsular development. Conversely, experimental significantly decreased over time, from 16% before starvation to 0% during starvation. Experimental were significantly less than controls on days 35-37. After starvation, percent capsular development increased to 32%.

**Juvenile Size.** Juveniles produced from control nudibranchs were generally larger than those produced by experimental nudibranchs (Figure 4.6C, Table 4.2).
Controls did not vary over time. During starvation, juveniles produced by experimental nudibranchs steadily declined in size and were significantly smaller than controls on day 36. Size of juveniles produced by experimental nudibranchs tended to increase again after food was returned.

**Discussion**

*Tenellia adspersa* is a generalist, and feeds on a variety of hydroids that are ephemeral in time and space (Clark, 1975; Harris and Irons, 1982). Due to the rapid changes in the distribution and abundance of these hydroids within the Great Bay Estuary (Chapter One) it is possible that a population of *Tenellia* living on a colony could exhaust its food supply within one generation. As observed here, stress caused by reduced food affected both the adults and their progeny.

After the onset of maturity, the daily number of eggs produced by well-fed *Tenellia* increased to a plateau while growth rate declined. Reproductive output was positively related to adult size. Larger nudibranchs produced more eggs per spawn and more eggs per day than smaller nudibranchs. This relationship has been demonstrated in other gastropods (e.g., Spight and Emlen, 1976) as well as in fishes, amphibians and reptiles (Stearns, 1992). Starvation had a significant impact on adult size and on reproduction. Starved adults decreased in size and produced substantially fewer spawn each with fewer eggs than well-fed adults. The reductions in growth and egg production both occurred within a day after the food was removed. Adult growth resumed within approximately one day after starvation ceased, while egg production did not increase until 3-4 days. These observations suggest that adult *Tenellia* possess few energy reserves to devote to reproduction and the majority of energy gained by feeding on *Cordylophora* is being channeled into reproduction. Calow and Wollhead (1977) observed a similar pattern in the turbellarian, *Dugesia lugubris*. Growth in *Dugesia* is sigmoidal, similar to that in *Tenellia*. When starved, egg capsule production continued for a short period of time after the initiation of starvation.
During this time adult size decreased rapidly until capsule production ceased, then adult size decreased more slowly. The sacoglossan opisthobranch, *Olea hansineensis* also has a short life cycle (3 months), high fecundity and a sigmoid growth curve (Chia and Skeel, 1973). As with *Tenellia*, prior to maturity most of the energy consumed by *Olea* was directed to growth. After maturity, the majority of energy was directed towards reproduction.

Adult starvation also affected the progeny. Variations in egg size and organic content in relation to adult nutritional stress have been demonstrated in a number of invertebrates (Bayne, *et al.*, 1975; Bayne, *et al.*, 1978; McKillup and Butler, 1979; Thompson, 1983). In the present study, egg size was significantly less in nudibranchs that had been starved for up to 5 days and investment per egg was significantly less in nudibranchs starved for up to 4 days. This decrease in size and investment was reversible, for when the nutritional stress was removed, egg size and egg weight increased to pre-starvation levels. For many marine invertebrates, a negative relationship exists between the number and size of eggs (Thorson, 1946, 1950; Emlet, *et al.*, 1987; Hermans, 1979; Bridges, 1993). Assuming that an organism has a finite amount of energy available for reproduction, an organism that produces small eggs will be able to produce more eggs compared to those that produce larger eggs (Vance, 1973; Smith and Fretwell, 1974; Stearns, 1976; Bridges, 1993). Upon maturity, *Tenellia* continues to feed on its hydroid prey with the majority of the energy being channeled to reproduction. Under well-fed conditions, larger eggs are produced. When the food source is removed or exhausted, smaller eggs were produced. These eggs were as much as 26% smaller in volume and 40% less in weight compared to eggs produced by fed adults. By decreasing egg size, *Tenellia* produces more eggs with its dwindling resource than could be produced by keeping egg size constant.

Helm *et al.* (1973) and Bayne *et al.* (1978) observed that eggs produced by stressed organisms tended to be of poorer quality than those produced by non-stressed
individuals. In contrast, eggs produced by starved Tenellia did not appear to differ from those produced by well-fed adults as was evident from comparable survival to metamorphosis. Further differences between offspring produced by fed and starved adults may also exist in post settlement growth and survival.

Changes in egg size and investment affected development time, development type and juvenile size. Under well-fed conditions, the eggs produced took longer to develop, and a portion of the eggs metamorphosed within the egg capsule. Juveniles produced from these eggs were larger. In contrast, all eggs produced by starved adults developed and were released as smaller pelagic lecithotrophic larvae. These larvae spent longer in the water column, potentially dispersing over greater distances. Thus, starvation did affect the early life history of the progeny. Hughes and Roberts (1980) observed that egg size was correlated with hatching size in the prosobranch, Littorina rudis. This may be a mechanism for survival within the estuary where hydroids are spatially patchy and temporally unpredictable. The results suggest that in a healthy hydroid colony, Tenellia would produce larger eggs that develop into larger juveniles, a proportion of which would metamorphose within the hydroid colony. This would also result in a quicker buildup of nudibranchs within the hydroid colony potentially leading to a faster demise of that hydroid colony. When the hydroid source is exhausted, Tenellia produces smaller offspring that disperse away from the hydroid colony.

Tenellia adspersa displayed developmental plasticity. Although this could be termed poecilocogy (presence of more than one developmental mode in the same species), Hoagland and Robertson (1988) have suggested that cases in which "...non-polymorphic, continuous variation in egg size, clutch size, or development time is not poecilocogy" (Pg. 110). Additionally, artificial delay of hatching has been observed in laboratory culture of spawn masses maintained in static cultures (i.e., no aeration, change of filtered seawater) as compared to spawn masses maintained in flowing seawater (Davis, 1967; Hurst, 1967; Harris, 1975; Harris et al., 1980; Todd, 1981;
Artificial delay has been suggested as an explanation in a number of reported cases of poecilogeny (Hoagland and Robertson, 1988). However, the phenomenon observed in *Tenellia* cannot simply be a case of artificial delay because spawn masses from the same individuals raised under identical conditions yielded two different developmental modes. Additionally, these observations demonstrate this plasticity in *Tenellia* is not a case of sibling species as has also been suggested for some cases of poecilogenous species.

There are other possible environmental components that could affect the egg matrix and in turn may ultimately affect development. Most opisthobranchs enclose their eggs within a gelatinous egg matrix that is secreted by the mucous gland and attached to the substratum (Ghiselin, 1966). Todd (1981) has suggested that the matrix protects the eggs from mechanical damage, infection, predation, osmotic stress, and chemical damage. Adult nutrition could play a role in the strength of this matrix. The estimate of investment per egg use here, measured not only the amount of material in the egg but the amount of material in the capsule and matrix. The decrease in investment observed during starvation may have been the result of a decrease in the amount of matrix. Thus, nutritional stress may cause nudibranchs to secrete a weaker matrix. A weaker matrix may erode at a faster rate, allowing veligers to hatch out at an earlier stage. Additionally, extrinsic biotic and abiotic factors, such as the effect of bacteria, ciliates, nematodes, harpactacticoid copepods and water movement also appear to be important in weakening the matrix and allowing escape of the larvae (Hurst, 1967; Harris, 1973; 1975; Harris, *et al.*, 1980; Todd, 1981).

Carroll and Kempf (1990) observed that all eggs of *Berghia verrucornis* hatch as pelagic lecithotrophic larvae when spawn masses were aerated, whereas some of the larvae metamorphose within the egg capsule when cultured in the absence of aeration. Eyster (1986) suggested that flowing water may provide more oxygen to developing embryos and increase rate of diffusion of embryonic wastes out of the egg capsule.
Low oxygen or high waste levels can inhibit development. Although this could have contributed to the developmental times observed in *Tenellia*, it cannot account for differences in development in spawn masses produced by the same individual under different nutritional states because all spawn were cultured under identical conditions. Harris (1973) observed that veligers of *Phestilla sibogae* and *P. melanobrachia* will not attempt to hatch from their capsules until the matrix has broken down sufficiently to release the capsules. Harris *et al.* (1980) also observed this for *Tenellia fuscata* (=*adspersa*), and suggested that variation in development may be the result of differential breakdown of the egg matrix caused by extrinsic biotic and abiotic factors. However, the present study clearly demonstrates that adult nutritional status had the greatest impact on developmental times and modes.

In conclusion, this study demonstrates *Tenellia* has a plastic developmental strategy. *Tenellia adspersa* responds to nutritional stress not only by a reduction in size and reproductive output but by altering egg size. This plastic life history strategy may be a mechanism for survival in an estuarine environment where hydroid prey is spatially patchy and temporally unpredictable.
Table 4.1. Life history characteristics of *Tenellia adspersa* under different food regimes. Control refers to nudibranchs that were provided an *ad libitum* diet of the hydroid *Cordylophora lacustris*. Experimental refers to nudibranchs that were also provided with an *ad libitum* diet of *Cordylophora* but were starved for 4 days (experiment one) or 7 days (experiment two). Size after starvation refers to the size of the experimental nudibranchs after starvation and the size of the control (fed) nudibranchs on the same day. Data presented as mean (sample size) ± 1 standard error. *P*-values are the results of t-tests comparing control and experimental treatments.

<table>
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<tr>
<th></th>
<th>Experiment #1, 25°C</th>
<th></th>
<th></th>
<th>p</th>
<th>Experiment #2, 20°C</th>
<th></th>
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<th>p</th>
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<tr>
<td></td>
<td>Control</td>
<td>Experimental</td>
<td>p</td>
<td>Control</td>
<td>Experimental</td>
<td>p</td>
<td></td>
<td></td>
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<tr>
<td>Egg Diameter (μm)</td>
<td>119.6 (12) 0.1</td>
<td>119.9 (12) 0.1</td>
<td>ns</td>
<td>119.8 (6) 0.2</td>
<td>119.9 (6) 0.2</td>
<td>ns</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Time to Metamorphosis (days)</td>
<td>8.17 (12) 0.26</td>
<td>8.55 (12) 0.26</td>
<td>ns</td>
<td>10.67 (6) 0.19</td>
<td>10.67 (6) 0.19</td>
<td>ns</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Size at Metamorphosis (μm)</td>
<td>214.5 (12) 3.6</td>
<td>217.7 (12) 5.8</td>
<td>ns</td>
<td>214.5 (6) 3.3</td>
<td>219.7 (6) 5.1</td>
<td>ns</td>
<td></td>
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<tr>
<td>Age at Maturity (days)</td>
<td>19.20 (10) 0.34</td>
<td>18.80 (10) 0.46</td>
<td>ns</td>
<td>28.40 (5) 0.22</td>
<td>28.67 (6) 0.19</td>
<td>ns</td>
<td></td>
<td></td>
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<tr>
<td>Size at Maturity (mm)</td>
<td>3.60 (10) 0.20</td>
<td>3.60 (10) 0.20</td>
<td>ns</td>
<td>4.25 (5) 0.29</td>
<td>4.04 (6) 0.21</td>
<td>ns</td>
<td></td>
<td></td>
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<tr>
<td>Maximum Length (mm)</td>
<td>6.01 (10) 0.34</td>
<td>5.06 (10) 0.32</td>
<td>ns</td>
<td>6.85 (5) 0.19</td>
<td>6.67 (6) 0.17</td>
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<td>Size after Starvation (mm)</td>
<td>6.32 (10) 0.48</td>
<td>3.61 (10) 0.24</td>
<td>***</td>
<td>6.04 (5) 0.41</td>
<td>3.72 (6) 0.13</td>
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<tr>
<td>Life Span (days)</td>
<td>27.00 (12) 2.82</td>
<td>32.4 (12) 2.72</td>
<td>ns</td>
<td>49.33 (6) 2.42</td>
<td>56.17 (6) 0.80</td>
<td>*</td>
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ns = not significant, * = p<0.05, *** p<0.001
Table 4.2. Results of two factor Analysis of Variance on adult reproduction, offspring size, and offspring development. Capsular period refers to time spent in capsule prior to hatching. Pelagic period refers to time from hatching to metamorphosis. N.A. = experiment not performed.

<table>
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<tr>
<th>Comparison</th>
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<th></th>
<th>Experiment Two</th>
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<td>Treat.</td>
<td>Day</td>
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<tr>
<td># of Spawn</td>
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<td>**</td>
<td>***</td>
<td>***</td>
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<tr>
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<tr>
<td># Eggs/Spawn</td>
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<td>***</td>
<td>**</td>
<td>***</td>
<td>***</td>
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<tr>
<td>Mean Egg Diameter</td>
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<td>***</td>
<td>***</td>
<td>***</td>
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<tr>
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<td>Mean Egg Weight</td>
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<tr>
<td>F-ratio</td>
<td>57.6</td>
<td>6.7</td>
<td>3.2</td>
<td>N.A.</td>
<td>***</td>
<td>***</td>
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<td>Juvenile Size</td>
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<tr>
<td>Capsular Period</td>
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<td>ns</td>
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<tr>
<td>% Metamorphosing</td>
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<td>*</td>
<td>ns</td>
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<tr>
<td>F-ratio</td>
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<td>1.7</td>
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<tr>
<td>% Capsular Dev.</td>
<td>**</td>
<td>***</td>
<td>***</td>
<td>ns</td>
<td></td>
<td>**</td>
<td></td>
</tr>
<tr>
<td>F-ratio</td>
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<td>12.1</td>
<td>6.6</td>
<td>5.6</td>
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ns = not significant, * = p<0.05, ** = p<0.001, *** = p<0.0001
Figure 4.1. Growth of Tenellia adspersa on two food regimes. Control nudibranchs were provided with an ad libitum diet of the hydroid, Cordylophora lacustris. Experimental nudibranchs were also fed the hydroid ad libitum except for a 4 day period of starvation. Inset is the number of nudibranchs alive each day.
Figure 4.2. Relationship between nudibranch size and reproductive output. A. Number of eggs laid per day. B. Average number of eggs per spawn.
Figure 4.3. Effect of starvation on average number of spawn masses laid (A) and average number of eggs per spawn (B). Reproductive output in two treatments over time. Period of starvation refers to 4 day period in which the experimental nudibranchs were without hydroid prey. Data from Experiment One. Bars = standard error of measurement. * = p<0.05 Tukey's HSD comparisons between treatments for each day.
Figure 4.4. Effect of starvation of mean egg size (A) and mean investment per egg (B), Experiment One. Bars = standard error of measurement. * = p < 0.05 Tukey's HSD comparisons between treatments for each day.
Figure 4.5. Effect of starvation on the progeny's total development time (A), capsular period (B) and pelagic period (C), Experiment One. Bars = standard error of measurement. * = p < 0.05 Tukey's HSD comparisons between treatments for each day.
Figure 4.6. Effect of starvation on mean egg diameter (A), percentage of eggs within a spawn undergoing capsular metamorphic development (B) and length of newly metamorphosed juveniles (C), Experiment Two. Bars = standard error of measurement. * = p < 0.05 Tukey's HSD comparisons between treatments for each day.

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CHAPTER V
THE RELATIONSHIP BETWEEN EGG SIZE, PARENTAL INVESTMENT AND OFFSPRING DEVELOPMENT

Introduction
One of the central paradigms in larval ecology relates the amount of parental investment to the amount of larval nutrition and developmental mode. Assuming that an organism has a finite amount of energy available for reproduction then there is a tradeoff between the size and number of eggs that can be produced (Thorson, 1946; 1950; Vance, 1973; Smith and Fretwell, 1974; Stearns, 1976; Emlet et al., 1987; Hermans, 1979; Bridges, 1993). Adults can invest little energy per egg, producing many small eggs which have a long pelagic period and obligate feeding in the plankton (planktothophy). At the other end of the continuum, adults can invest a large amount of energy per egg, producing few eggs which have a short or no pelagic period and subsisting on internal energy reserves (lecithotrophy). A number of empirical and theoretical studies have dealt with the evolution of developmental modes. The majority of these studies have used egg size as the predominant measure of parental investment (e.g., Vance, 1973). The advantage of using egg size is that it offers a non-destructive, easily obtainable measure.

In the majority of invertebrate groups studied, egg size is related to energy investment and developmental mode (Chia, 1976; Emlet, et al., 1987; Hadfield and Switzer-Dunlap, 1984; Rice, 1975; Thorson, 1950; Todd, 1981). For example, Strathmann and Vedder (1977) found a positive, non-linear relationship between egg size and energy investment in 10 invertebrates with small feeding larvae. Perron (1981) found highly significant positive relationships between egg size and development.
in six species of the gastropod *Conus*. However, McEdward and Coulter (1987) found no relationship between egg size and energy investment in populations of the echinoderm *Solaster stimpsoni*. Similarly, McEdward and Carson (1987) found no relationship in comparisons of egg size and energy investment in a single spawn of the echinoderm *Pteraster tesselatus*. Thompson (1983) found that in the urchin *Strongylocentrotus droebachiensis* organic content varied in response to nutritional stress while egg size did not. Variation in egg size and composition has been related to water temperature and latitude (Bayne, *et al.*, 1975; Bayne, *et al.*, 1978). These studies suggest that at taxonomic levels above species, egg size is correlated with investment, but at the intraspecific level egg size may be poorly correlated with energy investment. In addition, egg size may not accurately reflect changes in energy investment.

_Tenellia adspersa_ is a small (5-7 mm) estuarine aeolid nudibranch that is common to the Great Bay Estuary (NH). _Tenellia_ is a generalist and feeds on a variety of gymnoblastic and calyptoblastic hydroids (Clark, 1975). _Tenellia_ lays relatively small spawn masses containing 10-60 eggs. Chapter Four demonstrated that _Tenellia_ has a mixed developmental strategy; eggs either hatch as pelagic lecithotrophic larvae or metamorphosis within the egg capsule. The nutritional state of the adult affects both the size of eggs and the frequency at which the two developmental types are observed. Under well-fed conditions, adults produce eggs that are larger and 10-20% of the eggs undergo metamorphosis within the egg capsule. When adults are starved, the eggs produced are smaller in size, and all eggs hatch as pelagic lecithotrophic larvae.

The present study investigates the role of egg size in development with the specific purpose of evaluating the relationship between egg size, development time and developmental mode. Additionally, the amount of variation in egg size was explored.
Materials and Methods

The nudibranch, *Tenellia adspersa*, and the hydroid, *Cordylophora lacustris*, were collected from floating docks in the Great Bay Estuary, New Hampshire in 1991 and have been maintained in the laboratory as stock cultures. A description and details of the collection and maintenance of cultures appear in Chapter 2. For this study, 5 pairs of adult nudibranchs were isolated from the stock culture and maintained at 20°C and 20%/oo salinity with daily water changes. Spawn masses were collected and raised to metamorphosis. Twenty-four juvenile nudibranchs were isolated from the f1 progeny and maintained in separate stacking dishes. These nudibranchs were paired for 4 hours each day to allow copulation. They were provided with an *ad libitum* diet of *C. lacustris*. Beginning 3 days post-maturity the nudibranchs were starved for 7 days.

Under well-fed conditions, *Tenellia* produces 3-5 spawn per day with 10-60 eggs per spawn. I removed all spawn from each nudibranch daily and counted the number of eggs per spawn. One spawn per day was rinsed in 3% ammonium formate, dried at 80°C for 48 hours to constant weight and weighed using a Cahn Microbalance. Another spawn per day was also rinsed in 3% ammonium formate and the eggs carefully removed from the spawn matrix. These eggs were dried at 80°C for 48 hours to constant weight and weighed using a Cahn Microbalance.

Total investment per egg was calculated by dividing the weight of the spawn by the number of eggs. This represents the parental investment in egg, egg capsule and spawn matrix for each egg. Offspring investment was calculated by dividing the weight of eggs by the number of eggs and represents the parental investment in egg and capsule alone.

The remaining 1-3 spawn were cultured at 20°C and 20%/oo salinity with daily water changes to determine survivorship, development type and juvenile size. All cultured spawn were videotaped daily for 30 seconds at 40x through a Nikon microscope. The videotaped images were transferred to computer using NIH Imaging.
vers. 1.57. The eggs remain in their relative positions during development, allowing individual eggs to be followed to hatching and metamorphosis. All eggs within a spawn could thus be categorized as either undeveloped (no visible signs of development), pelagic lecithotrophic or capsular metamorphic development. I also recorded the diameter of each egg within a spawn to determine if egg size varied with development type.

Statistical analyses were performed using SYSTAT (SYSTAT Inc., Evenston, IL). A comparison of egg sizes between the two developmental types was accomplished using data from the first spawn from each individual in a two-factor analysis of variance model (ANOVA) with development type as one factor and adult as the second. Least-squares linear regression analysis was used to compare egg size with juvenile size, percent hatching and metamorphosing. Proportional data was arc-sine transformed prior to analysis. To compare the functional relationships between total investment and parental investment versus egg size, I used a Model II reduced major axis (RMA) regression model (LaBarbera, 1989). Egg size was calculated as the cube of the radius to linearize the data (Neefus, Personal communication). The slope of the RMA regression was calculated as the slope of the model I least-squares regression divided by the correlation coefficient (LaBarbera, 1989). 95% confidence limits were calculated following methods of McArdle (1987). The y-intercept was calculated using standard methods of Sokal and Rohlf (1981). Slopes of the two lines were compared using an approximate t-test (McArdle, 1988). Means ± standard errors are presented throughout.

Results

Developmental Mode and Egg Size

Seventy-nine spawn were collected and videotaped over the observation period. Of these, 23 contained clear images at hatching to classify the eggs. The remaining images could not be used because either the matrix had completely disintegrated or at
some point development became abnormal and the eggs within the spawn died. Figure 5.1 illustrates videotaped images of the same spawn on three successive days. Note that the eggs remained in their relative positions for the ten day period such that each image could be overlaid and the fate of each egg within that spawn determined. On Day 0 (shortly after egg laying) the spawn matrix was very transparent, clean and rubbery, making it resistant to breakage (Figure 5.1A). By day 5, a thin film of bacteria and detrital material had built up on the outside of the matrix (Figure 5.1B). On Day 10 this detrital film had increased and the matrix was less rubbery and more delicate (Figure 5.1C). Four swimming veliger larvae were observed in the dish containing this spawn. Note the newly metamorphosed juvenile right next to its discarded larval shell.

Comparisons between the diameters of the two developmental types using the first spawn from each individual revealed that the size of eggs hatching as juveniles was significantly larger than those hatching as swimming larvae for all adults tested (Two-Factor ANOVA: $F_{1,46} = 78.84$, $p < 0.0001$) (Figure 5.2). Mean egg size for juveniles was $125.16 \pm 1.23 \mu m$ and $109.69 \pm 1.23 \mu m$ for swimming larvae. All adults produced eggs of similar size (Two-Factor ANOVA: $F_{5,46} = 2.16$, $p = 0.075$) and there was no interaction between adult and development type (Two-Factor ANOVA: $F_{5,46} = 1.71$, $p = 0.150$).

**Egg Distribution**

The mean size of eggs hatching as juveniles was larger than eggs hatching as swimming larvae (Table 5.1) (Figure 5.3A). The coefficient of variation was the same for both development types. The distribution of eggs hatching as swimming larvae was significantly different from normal (Kolmogorov-Smirnov: $D = 0.082$, $n = 198$, $p < 0.01$). $G_1$ and $G_2$ statistics indicate that the distribution was broader than a normal distribution and skewed to the left, respectively (Table 5.1). The distribution of eggs hatching as juveniles was not different from a normal distribution (Kolmogorov-
Smirnov: $D = 0.073$, $n = 68$, $p = 0.46$) although it was slightly narrower than a normal distribution.

Cumulative percentages of each developmental type with respect to egg size revealed a transition from pelagic lecithotrophic development and capsular metamorphic development that occurred around $124 \mu m$ (Figure 5.3B).

**Egg Size Correlation**

Egg size was positively related to total investment and offspring investment (Figure 5.4, Table 5.2). The slopes of these two regressions were not significantly different from each other (adjusted $t = 0.89$, 35 d.f.) indicating that *Tenellia* invests a constant proportion into the matrix as in the egg regardless of egg size.

Juvenile size was positively related to egg size ($t = 8.46$, $n = 98$, $p < 0.0001$, $r = 0.654$) (Figure 5.5). Egg size accounted for 42.1% of the variability in juvenile size.

**Discussion**

Egg size has been related to energy investment and developmental mode in a number of invertebrates (e.g., Thorson, 1950; Rice, 1975; Chia, 1976; Strathmann and Vedder, 1977; Todd, 1981; Emlet et al., 1987; Hadfield and Switzer-Dunlap, 1984). The majority of these studies compared egg size, energy investment and developmental mode among species. However, as McEdward and Carson (1987) pointed out, natural selection operates on intraspecific variation in maternal investment and not between species. Egg size has been shown to be under genetic control at least in echinoderms (e.g., Lessios, 1990). Very few studies have compared egg size and energy content at the intraspecific level. McEdward and Coulter (1987) compared egg size and energy content in populations of the echinoderm, *Solaster stimpsoni* and did not find any relationship. McEdward and Carson (1987) also found no relationship in comparison of eggs from a single spawn of the echinoderm, *Pteraster tessulatus*. 

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Some of the variation in egg size can be the result of changes in energy investment that are not reflected in egg size. For example, Thompson (1983) found that eggs produced by the echinoderm, *Strongylocentrotus droebachiensis* fed on different food rations did not differ in size but did predictably differ in the amount of energy in the eggs. Egg size and/or energy investment also varies in relation to water temperature, latitude or adult age (Bayne, *et al.*, 1975; Bayne *et al.*, 1978; Thompson, 1983; Nakaoka and Matsui, 1994).

The present study demonstrates a significant relationship between parental investment, egg size and developmental mode in the nudibranch, *Tenellia adspersa*. Eggs hatching as juveniles were larger than those hatching as swimming larvae. Positive relationships were found between egg size, parental investment and juvenile size. However, the results also suggest that there are other factors affecting egg size and investment.

Development can be affected by the number and location of eggs within a spawn (Chaffee and Strathmann, 1984; Strathmann and Chaffee, 1984). Chaffee and Strathmann (1984) found that embryos deep within a gelatinous spawn mass developed more slowly than embryos near the surface of the mass. Development time and developmental mode are affected by the differential breakdown of the spawn mass. Harris (1973) and Harris *et al.* (1980) observed that veligers of the nudibranchs *Phestilla sibogae, P. melanobrachia, Aeolidia papillosa* and *Tenellia adspersa* will not attempt to hatch from their egg capsules until the matrix has sufficiently broken to release the capsules. Organisms such as bacteria, ciliates, nematodes and harpacticoid copepods may be important in weakening the gelatinous matrix and allowing escape of larvae (Hurst, 1967; Harris, 1973; Harris, 1975; Harris, *et al.*, 1980; Todd, 1981). Water motion is also important in the timing of hatching. Eyster (1986) suggested that flowing water may provide more oxygen or increase the diffusion of embryonic wastes out of the egg capsules, thereby decreasing development time. This has been observed...
in spawn masses raised under static laboratory culture conditions (no aeration and daily change of filtered seawater) when compared to spawn masses maintained in flowing seawater (Davis, 1967; Hurst, 1967; Harris, 1975; Harris, et al., 1980; Todd and Doyle, 1981). Temperature and salinity have also been shown to have an effect on development time (e.g., Hagerman, 1970; Harris, et al., 1980; Todd and Doyle, 1981).

The use of egg size as a measure of investment potentially can underestimate parental investment. The eggs of most opisthobranchs are enclosed within a gelatinous spawn mass, secreted by the mucous gland and attached to the substratum (Ghiselin, 1966). This gelatinous material can be very substantial and potentially costly to produce. Stickle (1973) found that in the gastropod, *Thais lamellosa* extra-embryonic capsular material accounted for up to 45% of the biomass lost from the body during spawning. Perron (1981) found in the gastropod, *Conus*, that large, slowly developing eggs had more elaborate extra-embryonic structures than smaller, more quickly developing ones. Using egg size as a measure of investment ignores this investment in extra-embryonic structures and may therefore underestimate parental investment and lead to a poor correlation between development and investment. The present study demonstrated that *Tenellia* invested a constant proportion of material into the spawn matrix as in the egg itself. A comparison of the two regressions suggested that investment in matrix accounted for at least one third of the total allocation to the spawn.

In conclusion, a relationship between egg size and investment (measured as dry weight) was found within a population of the nudibranch, *Tenellia adspersa*. However, this study also suggests that other factors also play a role. Because most opisthobranchs secrete a gelatinous spawn matrix around the eggs, egg size as a measure may underestimate maternal investment and give a weaker relationship between egg size and maternal investment.
Table 5.1. Distribution of egg sizes by development type for eggs produced by *Tenellia adspersa*. Swimming larvae refers to embryos that hatched as pelagic lecithotrophic larvae and juvenile refers to embryos that metamorphosed in the egg capsule and hatched as benthic juveniles.

<table>
<thead>
<tr>
<th></th>
<th>Swimming Larvae</th>
<th>Juvenile</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean egg Diameter (μm)</td>
<td>115.04</td>
<td>128.02</td>
</tr>
<tr>
<td>Range (μm)</td>
<td>90.00-131.57</td>
<td>103.32-150.79</td>
</tr>
<tr>
<td>Median (μm)</td>
<td>116.17</td>
<td>126.96</td>
</tr>
<tr>
<td>Sample Size</td>
<td>198</td>
<td>68</td>
</tr>
<tr>
<td>Coefficient of Variation</td>
<td>0.0717</td>
<td>0.0715</td>
</tr>
<tr>
<td>Skewness (G₁)</td>
<td>-0.482</td>
<td>0.063</td>
</tr>
<tr>
<td>Kurtosis (G₂)</td>
<td>-0.303</td>
<td>0.364</td>
</tr>
</tbody>
</table>
Table 5.2. Regression coefficients for reduced major axes regressions of total investment and offspring investment versus egg size.

<table>
<thead>
<tr>
<th></th>
<th>Total Investment</th>
<th>Offspring Investment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample Size</td>
<td>48</td>
<td>29</td>
</tr>
<tr>
<td>Correlation coeff</td>
<td>0.79</td>
<td>0.78</td>
</tr>
<tr>
<td>RMA Slope (95% CI)</td>
<td>1.58 (1.43-1.79)</td>
<td>2.13 (1.95-2.43)</td>
</tr>
<tr>
<td>RMA Intercept (95% CI)</td>
<td>0.31 (0.27-0.34)</td>
<td>-0.05 (-0.12-0.00)</td>
</tr>
</tbody>
</table>
Figure 5.1. VHS video images of the same spawn on three consecutive days. A. Spawn on Day 0. B. Spawn on Day 5. C. Spawn on Day 10. Scale bar in all three = 100 μm.
Figure 5.2. Mean egg diameter for eggs hatching as larvae and juveniles in six adults. Bars = Standard error of the means (n = 6).
Figure 5.3. Distribution of egg diameters for two developmental modes. A. Frequency histogram of egg diameters. "Larvae" and "Juvenile" refers to mean egg diameters for the two modes. B. Cumulative frequencies for the two developmental modes.
Figure 5.4. The relation between total investment (weight of egg plus matrix) and offspring investment (weight of egg only) versus egg size. Egg size measured as the cube of the radius $\times 10^{-3}$. Model II reduced major axis regression is fitted to both sets of data.
Figure 5.5. Juvenile size at metamorphosis versus egg diameter.
CHAPTER VI
THE INFLUENCE OF ADULT NUTRITIONAL CONDITION ON OFFSPRING GROWTH AND SURVIVAL.

Introduction
In the majority of invertebrate groups studied, egg size is associated with energy investment, development time and developmental mode (Thorson, 1946; 1950; Rice, 1975; Chia, 1976; Strathmann and Veddar, 1977; Todd, 1981; Hadfield and Switzer-Dunlap, 1984; Emlet et al., 1987). However, a number of studies have observed large amounts of variation in egg size and investment within a species and even within a single spawn (Bayne et al., 1978; Turner and Lawrence, 1979; Sinervo et al., 1984; McClintock and Pearse, 1986; McEdward and Carson, 1987; Lambert and Todd, 1994). Some of this variation can be accounted for by variation in maternal investment due to differences in adult nutritional status (e.g., Bayne et al., 1975; 1978; McKillup and Butler, 1979; Thompson, 1983; George, 1994). This variation in maternal investment has been shown to affect larval survivorship, size at settlement and juvenile growth, juvenile survival and behavior in a variety of organisms, including: insects (e.g., Wellington, 1965; Capinera and Barbosa, 1976; Leonard, 1970), gastropod mollusks (e.g., Hughes and Roberts, 1980), bivalve mollusks (e.g., Helm et al., 1973; Bayne et al., 1978), echinoderms (e.g., Sinervo et al., 1984; Sinervo and McEdward, 1988; George, 1994) and amphibians (e.g., Kaplan, 1980).

Tenellia adspersa is a small (5-7 mm) aeolid nudibranch common in New England estuarine environments (Clark, 1975). It has a subannual life cycle with a short generation time (2-3 weeks) and produces 3-5 spawn per day with 25-50 eggs per spawn (Chapter 2). Chapter 4 demonstrated that Tenellia has a plastic developmental
strategy; eggs from the same spawn produce veliger larvae that either hatch as pelagic lecithotrophic larvae or metamorphose within the capsule and hatching as benthic juveniles. This strategy is dependent on egg size; eggs larger than 125 μm metamorphose within the capsule (see Chapter Five). Adult nutritional state affects the size of the eggs and the frequency at which the two developmental modes are expressed. Under well-fed conditions, a portion of the eggs within a spawn undergo capsular metamorphic development and hatch as benthic juveniles. Whereas, under starvation conditions, all of the eggs within a spawn hatch as pelagic lecithotrophic veligers. In addition, juveniles produced from starved adults are smaller than those produced from well-fed adults.

*Tenellia* feeds on a variety of hydroids that are spatially and temporally unpredictable (Chapter One). Due to the growth and feeding characteristics of *Tenellia*, nudibranchs could exhaust their food resources within one generation (Harris, *et al.*, 1980; Chapter Two). This suggests that the environmental conditions in which *Tenellia* survives and reproduces are highly unpredictable, and as a consequence of the developmental strategy, offspring will vary in their development and size at metamorphosis. The present study investigated whether offspring differed in growth and survival in relation to adult nutritional regime. In other words, is there a cost to producing smaller offspring during nutritional stress.

**Materials and Methods**

The hydroid, *Cordylophora lacustris* was collected in September 1991 from a floating dock on the Oyster River, Durham, New Hampshire (70°55'N, 43°08'W). The nudibranch, *Tenellia adspersa* was collected in October 1991 from a floating dock on the Squamscott River, New Hampshire (70°56'N, 43°02'W). Both were maintained as stock cultures at 25°C and 20°/oo salinity following methods outlined in Chapter Three.
The experiment consisted of 6 pairs of adult nudibranchs collected as spawn from the stock cultures. These nudibranchs were maintained at 20°C and 20°/oo salinity and fed an *ad libitum* amount of *Cordylophora* until 3 days after maturity. At this time the hydroid was removed and the nudibranchs starved. Beginning at maturity, spawn masses were collected daily and the number of eggs per spawn was counted at 60x using a Wild dissecting microscope. The diameter of all uncleaved eggs was measured using at 40x using a Nikon compound microscope. These spawn were then cultured at 20°C and 20°/oo salinity with daily water changes. Metamorphosed juveniles were provided with and *ad libitum* amount of *Cordylophora*. Offspring were followed for approximately 22 days, at which time hydroid prey ran out. The number of surviving individuals was counted daily. Beginning at metamorphosis, the size of each juvenile was measured daily at 60x using a Wild dissecting microscope.

This experiment was repeated once. However, due to poor laboratory water conditions, the second replicate was performed at 20°C and 30°/oo salinity using the same procedures as above.

Statistical analyses were accomplished using the SYSTAT statistical program, vers. 5.03 (SYSTAT Inc., Evanston, IL). An analysis of variance (ANOVA) model was used to compare egg size, development and juvenile size between offspring from the different treatments (well-fed versus starved). Following a significant F-test, Dunnett’s test was employed to compare the control group (offspring from well-fed adults) with offspring from starved adults (Zar, 1996).

Juveniles grow exponentially in size until maturity is reached (Chapter Two). Therefore, nudibranch size was transformed using a natural log (ln (x)) (Krebs, 1989) and the resulting data analyzed using least-squares linear regression analysis. Homogeneity of slopes was tested using an F-test. Dunnett’s test was used to compare the slopes of offspring from well-fed adults (control) with those of offspring from starved adults (Zar, 1996). Means ± standard errors are presented throughout.
Results

**Egg Diameter.**

Mean diameter of eggs from well-fed adults was higher in the second replicate compared to the first (t-test: $t_4 = 3.26$, $P < 0.05$). Mean diameter of eggs was 122.6 ± 1.7μm (n = 6 spawn) in replicate 1 and 128.8 ± 0.9μm (n = 6 spawn) in replicate 2.

In replicate 1, starvation resulted in a significant reduction in egg size ($F_{5,30} = 9.20$, $P < 0.0001$) (Figure 5.1). Egg size decreased from 122.6 ± 1.5 μm (n = 6 spawn) for well-fed adults to 115.1 ± 1.3 μm (n = 6 spawn) for adults starved 5 days. Starvation had a similar effect on egg size in replicate 2 ($F_{4,15} = 91.88$, $P < 0.0001$). Eggs decreased in size from 128.8 ± 0.7 μm (n = 6 spawn) for well-fed adults to 105.5 ± 0.6 (n = 3 spawn) for adults starved 4 days.

**Development.**

A significant decrease of approximately 3 days was observed in the time spent in the capsule (time to hatching) as a result of adult starvation (Figure 5.2). This was observed in both replicate 1 ($F_{5,30} = 4.71$, $P < 0.01$) and replicate 2 ($F_{4,15} = 91.88$, $P < 0.0001$). A decrease in total development time (time to metamorphosis) in response to starvation was also observed in replicate 1 ($F_{5,30} = 4.71$, $P < 0.01$). However, no change in total development time was observed in replicate 2 ($F_{4,15} = 2.37$, $P = 0.07$).

Offspring from well-fed adults metamorphosed into juveniles 240-250 μm in size (replicate 1: 237 ± 4μm, n = 6; replicate 2: 248 ± 3 μm, n = 16). Size of the newly metamorphosed juveniles declined in response to adult starvation in both replicate 1 ($F_{5,30} = 27.18$, $P < 0.0001$) and in replicate 2 ($F_{4,42} = 17.85$, $P < 0.0001$) (Figure 5.3). Offspring from adults starved 4-5 days metamorphosed into juveniles 180-190 μm in size (replicate 1: 188 ± 3μm, n = 6; replicate 2: 181 ± 5μm, n = 6).
**Juvenile Growth.**

Juveniles in replicate 1 grew exponentially in size (Figure 5.4A). Regressions of log-transformed sizes against age provided a very good fit to the data with positive slopes for all treatments (Table 5.1). The slopes of these regressions were significantly different from each other ($F_{5,218} = 3.57, P < 0.005$) (Figure 5.5A). However, Dunnett's test revealed that offspring from adults that had been starved for one day grew at a faster rate than offspring from well-fed adults. Offspring from adults starved longer than one day grew at the same rate as those offspring from well-fed adults.

Juveniles in replicate 2 also grew exponentially in size (Figure 5.4B), with the exception of offspring from adults starved 2 days. These offspring failed to grow and all died after 12 days post-metamorphosis. Consequently, this treatment was removed from the linear regression analysis. Regressions of log-transformed sizes against age revealed positive growth in all of the remaining treatments (Table 5.1). Slopes were not homogeneous ($F_{3,161} = 9.74, P < 0.0001$) (Figure 5.5B). Offspring from adults starved one and three days grew at a slower rate, compared to offspring from well-fed adults. However, offspring from adults starved four days grew at the same rate as those from well-fed adults.

**Juvenile Survival.**

Offspring survival was higher in replicate 1 compared to replicate 2 for all treatments (Figure 5.6). In all cases, the critical survival period appeared to be between 6 and 10 days when hatching and metamorphosis occurred.

Offspring from starved adults generally had a higher survival to hatching in replicate 1 ($F_{5,24} = 3.46, P < 0.05$) (Figure 5.6A). Survival increased from $0.35 \pm 0.06$ ($n = 5$) in offspring from well-fed adults to $0.73 \pm 0.09$ ($n = 5$) in offspring from adults starved 5 days. Dunnett's test identified that offspring from adults starved for 5 days had higher survival to hatching compared to offspring from well-fed adults.
No differences in survival to metamorphosis were observed between treatments in replicate 1 ($F_{5,23} = 1.11, P = 0.38$). Average survival to metamorphosis for all treatments was $0.35 \pm 0.03$ (n = 29). Additionally, in this replicate, no differences were observed in survival to the end of the experiment were observed between treatments ($F_{5,24} = 1.69, P = 0.18$). Average survival at the end of the experiment was $0.11 \pm 0.02$ (n = 30) for all treatments.

In replicate 2, survival to hatching did not significantly vary ($F_{4,23} = 1.53$, P = 0.23) and averaged $0.39 \pm 0.05$ (n = 28) for all treatments (Figure 5.7B). Survival to metamorphosis also did not vary ($F_{4,22} = 0.86$, P = 0.50) and averaged $0.18 \pm 0.03$ (n = 27) for all treatments. Survival at the end of the experiment did not vary ($F_{4,23} = 0.93$, P = 0.46) and averaged $0.03 \pm 0.01$ (n = 28) for all treatment groups.

**Discussion.**

Egg size and maternal investment can have an important impact on offspring growth and survival. For example, hatching success and naupliar survival in the copepod *Calanus helgolandicus* increased with increasing egg size and amount of protein, carbohydrate and lipid in the egg (Guisande and Harris, 1995). Egg size in the seed beetle, *Callosobruchus maculatus* affected development and emergence of adults (Fox, 1993; Fox, 1994). In the present study, egg size in the nudibranch, *Tenellia adspersa*, had a significant effect on development and size at metamorphosis but not on survival and growth of the post-metamorphic juveniles.

*Tenellia adspersa* has a plastic developmental strategy; developing embryos from the same spawn hatch as either pelagic lecithotrophic larvae or metamorphose within the egg capsule and hatch as benthic juveniles, depending on maternal investment. Adult nutritional state affects the size of the eggs and the frequency of the two developmental modes. Adult starvation had a significant and dramatic effect on egg size, offspring development and size at metamorphosis. Eggs from starved adults
were smaller, and all developed into pelagic lecithotrophic larvae that hatched earlier, spent longer in the plankton and metamorphosed into smaller juveniles. The previous chapter fully documented these changes and suggested that this plastic developmental strategy is a "bet-hedging" strategy allowing *Tenellia* to survive and reproduce in an environment where hydroid prey is patchily distributed and temporally unpredictable. Kaplan and Cooper (1984) proposed that if different egg sizes are favored under different conditions that stocastically vary among generations, then within-individual variation in egg size could represent a diversified bet-hedging strategy. Bet-hedging organisms sacrifice their potential fitness in order to reduce the probability of complete failure (Cohen, 1966; Seger and Brockman, 1987). This strategy involves a trade-off between the mean and variance of fitness (Phillipi and Seger, 1989). In stochastically, temporally fluctuating environments, fitness should be measured as the geometric mean of the finite rate of increase (Roff, 1992). The geometric mean decreases as the numbers being averaged become more variable (Phillipi and Seger, 1989). In diversified bet-hedging, the geometric mean fitness over generations is increased by decreasing the variance of the mean-within generation fitness.

The present study observed no differences in survival to metamorphosis between offspring from well-fed adults and offspring from adults that were starved for a period of up to 5 days. Offspring from starved adults did have a higher survival to hatching. However, this was only observed in replicate 1. Potential mortality due to predation, dispersal away from suitable settlement sites (i.e., out of the estuary and away from hydroid prey) or lack of a suitable settlement site was not accounted for in this laboratory study because pieces of hydroid were always available to newly hatched larvae. Potential mortality for pelagic larvae could be quite high compared to mortality of benthic juveniles. Yet, even if the survival for dispersing offspring was lower than non-dispersing offspring, when food resources are exhausted the dispersing offspring should have better survival than the non-dispersing offspring.
At 20\(^{\circ}\) salinity, offspring from starved adults grew at rates similar to offspring from well-fed adults. This was also the case for one starvation group raised at 30\(^{\circ}\) salinity, while the other groups grew at a slower rate compared to offspring from well-fed adults. This suggests that although offspring from nutritionally stressed adults metamorphose into smaller juveniles, the smaller juveniles were able to grow as fast and reach comparable sizes as the larger juveniles from well-fed adults.

In *Tenellia adspersa*, individuals might be sacrificing potential fitness by exhibiting more than one developmental mode (pelagic larvae or benthic juveniles). As noted above, larval mortality for the pelagic larvae may be higher than for the benthic juveniles. In a favorable environment, where the hydroid colonies are large and healthy, potential fitness would therefore be higher if *Tenellia* produced only benthic juveniles. However, hydroid colonies are temporally unpredictable and a particular colony may be completely wiped out within a single nudibranch generation, depending on the number of nudibranchs in the colony and on the hydroid colony size (Chapters One and Two). So, the risk of complete failure to reproduce is much higher. The variance in fitness measured over generations would also be higher because in some generations there would be enough hydroid prey to allow for maximum reproduction and the highest possible fitness, while in other generations hydroid prey could run out before *Tenellia* finishes reproduction or even before it matures. Although fewer offspring would potentially survive, by producing pelagic larvae and benthic juveniles, *Tenellia* decreases the probability of complete failure and also decreases the variance in fitness. The decrease in variance also increases the geometric mean of fitness.

The present study demonstrates that juvenile growth and survival at the lower salinity was variable but with no pattern to this variation. Although, offspring from starved adults metamorphosed into smaller juveniles, these juveniles grew as fast and survived as well as offspring from well-fed adults. This fits the hypothesis of a bet-
hedging strategy. If growth and survival of smaller offspring were lower then one would expect selection to favor larger eggs and offspring size would increase.

Examples are found in other invertebrate systems. Egg provisioning in the gypsy moth, *Lymantria dispar* represents a bet-hedging strategy in response to an unpredictable environment (Rossiter, 1991). Some freshwater pisidiids (e.g., *Sphaerium occidentale, Musculium portumeium* and *M. securis*) have a quasi-iteroparous reproductive strategy that has been suggested as a bet-hedging strategy (Mackie, 1984).

The poor growth of offspring from nutritionally stressed adults observed at higher salinity suggests that other environmental factors may also play a role. Conditions in the second replicate were not as optimal as in the first replicate. In addition to higher salinity, the prey source, *Cordylophora*, was not as healthy and did not survive long at this higher salinity (see Chapter Two). It is possible that the importance of adult nutritional state to offspring growth and survivorship depends on the favorability of environmental. Under stressful conditions adult nutritional state could be more important to the offspring and adult starvation would incur a greater cost to the offspring than under more benign conditions.

In conclusion, this study demonstrates that although adult nutritional state affected egg size and development of offspring it did not affect juvenile growth and survival, suggesting that any cost to the offspring associated with adult starvation is short-term and confined to the pre-metamorphic stages. It is further suggested that this plastic developmental strategy, where both pelagic lecithotrophic larvae and benthic juveniles are produced in a single spawn, is a bet-hedging mechanism allowing *Tenellia* to survive and reproduce in a temporally and spatially unpredictable and potentially stressful environment.
Table 6.1. Results of least-squares regression analysis of juvenile size on age. Juvenile size was transformed using the natural log (x). Replicate One performed at 20°C and 20°/00 salinity, replicate two performed at 20°C and 30°/00 salinity. Dunnett's test was used to compare the slopes of offspring from starved adults with those from adults that were well-fed. * = p < 0.05 at the α = 0.05 level.

<table>
<thead>
<tr>
<th>Days</th>
<th>Adults Starved</th>
<th>Correlation Coefficient (r)</th>
<th>Y-intercept</th>
<th>Slope</th>
<th>n</th>
<th>Dunnett's Test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Replicate One</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Zero days</td>
<td>0.97</td>
<td>5.10</td>
<td>0.20</td>
<td>51</td>
<td></td>
<td></td>
</tr>
<tr>
<td>One day</td>
<td>0.95</td>
<td>5.18</td>
<td>0.23</td>
<td>40</td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>Two days</td>
<td>0.96</td>
<td>5.18</td>
<td>0.19</td>
<td>53</td>
<td>ns</td>
<td></td>
</tr>
<tr>
<td>Three days</td>
<td>0.98</td>
<td>5.14</td>
<td>0.18</td>
<td>22</td>
<td>ns</td>
<td></td>
</tr>
<tr>
<td>Four days</td>
<td>0.99</td>
<td>4.97</td>
<td>0.21</td>
<td>18</td>
<td>ns</td>
<td></td>
</tr>
<tr>
<td>Five days</td>
<td>0.98</td>
<td>5.02</td>
<td>0.21</td>
<td>46</td>
<td>ns</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Replicate Two</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Zero days</td>
<td>0.98</td>
<td>5.11</td>
<td>0.19</td>
<td>62</td>
<td></td>
<td></td>
</tr>
<tr>
<td>One days</td>
<td>0.99</td>
<td>5.14</td>
<td>0.17</td>
<td>44</td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>Three days</td>
<td>0.99</td>
<td>5.09</td>
<td>0.17</td>
<td>29</td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>Four days</td>
<td>0.98</td>
<td>4.92</td>
<td>0.19</td>
<td>34</td>
<td>ns</td>
<td></td>
</tr>
</tbody>
</table>
Figure 6.1. Mean egg diameter of offspring produced from adults that were well-fed compared to adults that had been starved for 1 - 5 days. A. Experiment One, adults at 20°C 20°/oo salinity. B. Experiment Two, adults at 20°C 30°/oo salinity.
Figure 6.2. Time to hatching and to metamorphosis for offspring from adults that had been well-fed compared to adults starved for 1-5 days. A. Experiment One, adults at 20°C 20‰ salinity. B. Experiment Two, adults at 20°C 30‰ salinity.
Figure 6.3. Size at metamorphosis of offspring produced from adults that were well-fed compared to adults that had been starved for 1-5 days. A. Experiment One, adults at 20°C 20°/oo salinity. B. Experiment Two, adults at 20°C 30°/oo salinity.
Figure 6.4. Size versus age for offspring from adults starved 0-5 days. A. Experiment One, adults at 20°C 20°/oo salinity. B. Experiment Two, adults at 20°C 30°/oo salinity.
Figure 6.5. Growth rate of offspring from adults starved 0-5 days. Size was transformed using natural log of the size. Least squares linear regression applied to the data. A. Experiment One, adults at 20°C 20%/oo salinity. B. Experiment Two, adults at 20°C 30%/oo salinity.
Figure 6.6. Survival of offspring from adults starved 0-5 days. Survival is measured as survival to age \( x (L_x) \). A. Experiment One, adults at 20°C 20\%/oo salinity. B. Experiment Two, adults at 20°C 30\%/oo salinity.
Figure 6.7. Survival to hatching, metamorphosis and to the end of the experiment for offspring from adults starved 0-5 days. Survival is measured as survival to age x ($L_x$). A. Experiment One, adults at 20°C 20°/oo salinity. B. Experiment Two, adults at 20°C 25°/oo salinity.
GENERAL CONCLUSIONS

The aeolid, *Tenellia adspersa* is a small (5-7mm) nudibranch common in New England estuarine environments. *Tenellia* has a subannual life cycle with a lifespan of one to two months at ambient sea temperatures. *Tenellia* feeds on a variety of gymnoblastic and calyptoblastic hydroids. This dissertation has shown that *Tenellia* lives in an unpredictable environment where not only physical conditions (temperature and salinity) can change but hydroid prey distribution and abundance can change in as little time as a week. It has also been demonstrated that *Tenellia* can exhaust its food source within a single generation depending on initial hydroid colony size and salinity regime. *Tenellia* also varies its life history strategy (i.e., age and size at maturity) in response to the hydroid species it settles on.

*Tenellia adspersa* has a plastic developmental strategy. Eggs from the same spawn develop into veliger larvae that either hatch as pelagic lecithotrophic larvae or metamorphose within the capsule and hatch as benthic juveniles. Egg size is positively related to parental investment in *Tenellia*. This developmental strategy correlates with egg size (i.e., parental investment). eggs larger than 125 µm metamorphose into benthic juveniles. Adult nutritional state affects the size of the eggs and the frequency at which the two developmental modes are expressed. Under well-fed conditions, eggs are larger and 5-20% of the eggs within a spawn undergo capsular metamorphic development and hatch as benthic juveniles. Whereas, under starvation conditions, eggs are smaller and all eggs within a spawn hatch as pelagic lecithotrophic veligers. These smaller larvae spend longer in the plankton and metamorphose into smaller juveniles.

Although adult nutritional state affected egg size and offspring development, it did not affect juvenile growth and survival. The results suggest that any cost to the
offspring associated with adult starvation is confined to the premetamorphic stages. Given that *Tenellia* lives in an unpredictable environment, this developmental strategy where both pelagic lecithotrophic larvae and benthic juveniles are produced in a single spawn may represent a bet-hedging strategy. *Tenellia* is minimizing the chance of complete failure by producing both offspring that will remain with the hydroid colony and offspring that will disperse to another hydroid colony.

The research present in this dissertation has implications for mariculture. The research presented in Chapters Four and Six demonstrate that the adult nutritional regime can be important in determining the quality (i.e., larval development and survival) of the offspring. Maintenance of well-fed adult brood stocks would therefore be important in producing good quality offspring in sufficient numbers.

This research has also demonstrated the feasibility of culturing both a nudibranch and its hydroid prey in a low-cost, easy maintenance closed-culture system. In addition, metamorphic competency and induction are not critical elements. At present, the culture was maintained using natural seawater. However, very little additional work would be needed to adapt this system to use artificial seawater. *Tenellia adspersa* and its food source, *Cordylophora lacustris*, have been shown to be a good model system for use in studying predator-prey systems and life history studies because of the ease at which both can be cultured in lab, the fast generation time of the nudibranch, and short non-feeding larval stage.
LITERATURE CITED


Caswell, H. 1983. Phenotypic plasticity in life-history traits: demographic effects and evolutionary consequences. American Zoologist. 23:


Kaplan, R.H. 1980. The implications of ovum size variability for offspring fitness and clutch size within several populations of salamanders (Amblystoma). Evolution. 34: 51-64.


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Peterson, B. 1950. The relation between size of mother and number of eggs and young in some spiders and its significance for the evolution of size. Experimentia. 6: 96-98.


