DEIDRE MICHELLE GIBSON
Feeding Growth and Reproduction Rates of the Doliolid *Dolioletta gegenbauri*
Uljanin (Tunicata, Thaliacea)
(Under the Direction of GUSTAV ADOLF PAFFENHÖFER)

The objectives were to quantify feeding and growth of *Dolioletta gegenbauri* gonozooids and reproduction rates of *Dolioletta gegenbauri* phorozooids at 4 food concentrations, 3 food sizes, and 4 temperatures in order to determine their impact as grazers throughout the water column. Laboratory experiments were conducted at 16.5, 20, 23.5, and 26.5°C quantifying removal of a 50:50 volumetric concentration of *Thalassiosira weissflogii* and *Rhodomonas* sp. at four different food concentrations of 7 20, 60, 160, and 390 µg C · l^{-1}. Gonozooid clearance rates are similar at concentrations from 20 to 60 µg C · l^{-1}, and decrease as the food concentrations increase to 160, and 390 µg C · l^{-1}. The ingestion rates increase over a range of phytoplankton concentrations from 20 to 160 µg C · l^{-1}, then decrease when abnormally high concentrations of 390 µg C · l^{-1} are offered. Clearance and ingestion rates increase as temperature increases from 16.5 to 26.5° C. The exponential growth rates range from k = 0.2 - 0.7 with the lowest rates occurring at the highest food concentration. Growth rates increase with increasing temperature from K = 0.1-0.3/day at 16.5° C to 0.45-0.7 at 26.5° C. Phorozooid release rates increased at 20 $^{\circ}$C as concentration increases from 7 to 160 µg C/l. Release rates remained similar as the temperature increased from 16.5 to 26.5 $^{\circ}$C at a phytoplankton concentration of 60 µg C/l. Food concentration and temperature has an affect on phorozooid reproductive longevity, size of gonozooids released, and the amount of carbon released asexually. As *Dolioletta gegenbauri* gonozooids increased from 3.5 to 5.5 mm, their ability to ingest *R. alata* increased. Small and medium sized gonozooids can ingest large and small cells at similar rates at all concentrations, while at highest concentration of *R. alata* the larger gonozooid's clearance rates on the large diatom, reduced the grazing pressure on smaller cells. Doliolid feeding, growth and reproduction rates are a function of environmental food size and concentrations and temperatures, and imply that they can be important consumers in a changing neritic environment.

INDEX WORDS: *Dolioletta gegenbauri*, Gonozooid, Phorozooid, Feeding rate, Growth rate, Reproduction rate, Food concentration, Temperature

FEEDING, GROWTH AND REPRODUCTION RATES OF THE DOLIOLID, *DOLIOLETTA GEGENBAURI* ULJANIN (TUNICATA, THALIACEA)

by

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CHAPTER I

INTRODUCTION / LITERATURE REVIEW

Until two decades ago, planktonic copepods were seen as the major grazers on phytoplankton. This view changed when feeding and growth rates of protozoa were quantified. For example, during late spring to early summer in the North Atlantic Ocean, protozoa removed between 37 and 115% of the primary production per day (Burkill et al. 1993, Verity et al. 1993a) whereas copepods removed only 0.6 to 5.2% (Dam et al. 1993). Similar conditions were observed in the Subarctic Pacific Ocean (Miller 1993). It has been argued that salps, due to their high clearance and growth rates, can also remove significant amounts of the daily primary production (e.g. Harbison and Gilmer 1976). The apparent mismatch of relatively low clearance rates (e.g. Deibel 1982b, 1985b, Mullin 1983) and rather high growth rates (*Thalia democratica*, Heron 1972, Heron and Benham 1985), can not support independently determined growth rates. High abundances of thaliaceans, salps and doliolids, have usually been reported intermittently, and not continuously, in neritic regions (e.g. Braconnot 1963, 1971, French Mediterranean Coast; Berner 1967, California Current; Binet 1976, Ivory Coast; Hopkins and Torres, 1989, Antarctic Ocean). Thus, the impact of thaliacea on primary production has remained uncertain. The limited knowledge on sexual and asexual reproductive rates and their longevity has furthered this uncertainty.

Recent oceanographic observations on the southeastern continental shelf revealed that thaliacea, in particular doliolids, occurred frequently during winter, spring and summer (Deibel 1985a, Paffenhöfer et al. 1984, 1987, 1995), for periods of up to 4 weeks (Paffenhöfer and Lee 1987a). Doliolids were abundant on about 50% of cross-shelf cruises on the southeastern continental shelf between 31° and 32°N during the past three years (Paffenhöfer, unpubl.). Doliolids and salps occurred year-round off the Ivory Coast (Binet 1976). During spring, summer and fall, Esnal and Simone (1982) recorded doliolid nurses at 77% of all stations taken in the Caribbean and Gulf of Mexico.

Estimated *in situ* feeding rates of doliolids, derived from field abundances and sizes, and laboratory rates, ranged from clearing < 10% to >100% of the water per day (Deibel 1985a, Paffenhöfer et al. 1995). These high rates indicate that at certain times doliolids may significantly modify concentrations of phytoplankton and other particles.

However, the absence of information on functional responses of the various zooids (Deibel 1995), of asexual and sexual reproduction rates, and of environmental growth rates makes it impossible to characterize the effects which an assemblage of doliolids may have on their environment.

BACKGROUND

Life Cycle

Pelagic tunicates consist of two classes. The appendicularia are found circumglobally as are the thaliacea, except for estuaries. Salpidae and doliolidae are two of the three families representing the thaliacea, of which the latter usually occur only in neritic regions. Both reproduce sexually and asexually. The reproductive cycle of the doliolids consists of several types of zooids (Fig. 1). Gonozooids reproduce sexually through eggs and sperm. Larvae appear within 24 hrs after egg release and become an early 1 mm oozooid within 1 day. The oozooid develops a threadlike structure, the cadophore, which sprouts two small trophozooids, which feeds for the colony, five days after the oozooid appears. The number of trophozooids usually doubles every 24 hours, and by this time, the oozooid becomes a nurse at 2 to 3.5 mm. Small phorozooids appear on the cadophore by the time the nurse has at least eight trophozooids. Phorozooids can be released one day latter, and after four days the first signs of gonozooid buds appear on the phorozooids peduncle. Gonozooids are released at 2 to 3 mm, and become mature and release egg and sperm at > 6mm. The appearance of larvae and oozooids completes the life cycle in 20 days (Paffenhöfer and Gibson, 1999).

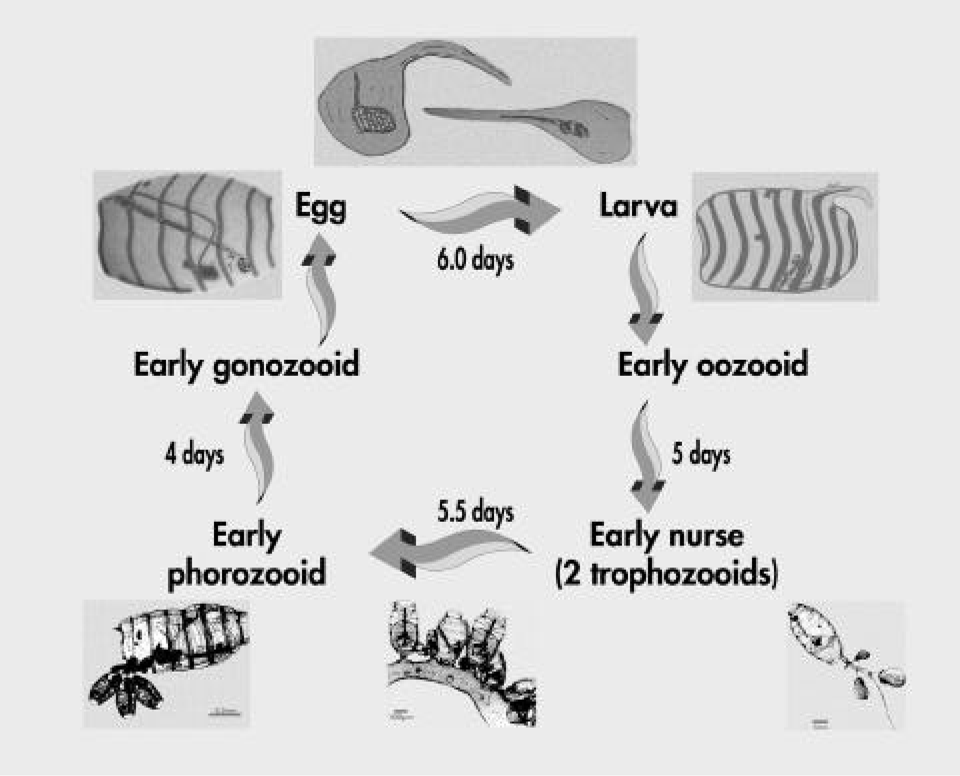

Fig. 1. Life cycle of *Dolioletta gegenbauri*.

Zoogeography

Doliolids have been *qualitatively* observed at temperatures from 3° to 28°C in Antarctic waters (Hopkins and Torres 1988), off South Africa (De Decker 1973), West Africa (Seguin and Ibanez 1974), off eastern Australia (Sheard 1965), off Peru (Timonin et al. 1979), off California (Mackas et al. 1991), in the Gulf of Mexico (Esnal and Simone 1982), in the equatorial Atlantic (Godeaux 1962) and in the North Sea (Lindley et al. 1990). They were *quantitatively* collected off Japan, China, India, Brazil, the Ivory Coast, California and on the southeastern shelf of the U.S. Concentrations were repeatedly registered in excess of 1,000 zooids m^{-3}. While many of these observations occurred during brief cruises and single transects, some studies allowed for repeated coverage of the same location or of the entire water masses. Paffenhöfer et al. (1987) observed high abundances of doliolids and development of a patch of doliolids mainly in the thermocline and cold deep layer where also phytoplankton was more abundant, and of the salp *Thalia democratica* in the warm upper mixed layer from June to August 1981. These patches eventually covered 9,300 km^2. Despite excess nitrate, no increases in phytoplankton concentrations occurred in the region covered by both patches, whereas phytoplankton concentrations increased shoreward of the patches (Paffenhöfer and Lee 1987a,b). Doliolids were also recorded abundantly during winter across the entire continental shelf off Charleston, South Carolina, over 10 days during January-February 1991 (Paffenhöfer et al. 1995). In the weakly stratified water column near the 40m isobath, doliolid larvae were most abundant in the deeper layer, the nurses in the middle of the water column, and phoro- and gonozooids in the upper layer.

Rates

Although doliolids have been found abundantly in numerous regions of the coastal ocean, data on rates of feeding, growth and reproduction are scarce. There are only three published studies on feeding rates, using different methodology. Deibel (1982b) quantified removal of algae over ~24h, Crocker et al. (1991) used radiolabel accumulation of bacteria or algae over periods of 1 to 5h, and Tebeau and Madin (1994) determined the number of microspheres which zooids had ingested during 5 to 15 min by counting the microspheres in the preserved animals' body. They obtained the highest feeding rates of those three studies probably due to gentle collection of the zooids, and higher temperatures. Crocker et al. (1991) obtained the lowest rates perhaps reflecting small container volumes, low temperatures, high food concentrations, and periods which were too long for radiolabel studies (see comment in Tebeau and Madin 1994). Deibel's (1982b) gonozooid rates were close to laboratory results on the feeding of *D. gegenbauri* indicate that clearance rates decrease with increasing concentrations of phytoplankton, (using a 50:50 mixture of diatoms and flagellates in terms of µg C · l^{-1}).

Deibel, 1982a published the only data on the rates of growth and reproduction. Phorozooid's of *D. gegenbauri* released on average 31 gonozooids at 20ºC and food concentrations of *Isochrysis galbana* and *Peridinium trochoideum* averaging 77 µg C · l^{1}. In preliminary studies *D. gegenbauri* phorozooids (n=9) produced on average 11.4 gonozooids · day^{-1} over an average of 11.8 days, with a maximum of 252 gonozooids produced per phorozooid over 20 days. Nurses with trophozooids produced phorozooids for up to 2.5 weeks. Daily exponential growth coefficients (k) ranged from 0.08-0.25 for gonozooids and 0.17-0.69 for phorozooids

(Deibel 1982a). These rates are below those observed by Heron (1972) for the salp *Thalia democratica* near 18-20ºC, and by Paffenhöfer (1976) for the appendicularian *Oikopleura dioica*, at 18ºC, in both cases exceeding k=1. These high growth rates could be attributed to offering recently collected natural prey, using large volume containers to limit wall effects (Heron had 50 l aquaria, Paffenhöfer up to 8-l-vessels), and Heron used *T. democratica* gently collected near the surface. It could also be that doliolids may be growing at a slower rate than co-occurring salps and oikopleurids.

This background information reveals that a sound assessment of the contribution of doliolids to consumption and production in neritic planktonic communities is not possible because of the paucity of rate data. We propose that previously perceived notion, that protozoa and copepods are the main consumers of phytoplankton in subtropical neritic regions may need to be amended due to high abundance and often occurrence of doliolids in conjunction with their high rates of feeding, growth, and reproduction.

OBJECTIVES

The overall goal of this study is to significantly enhance our knowledge on feeding, growth and reproduction of doliolids in order to be able to evaluate their contribution to the planktonic community as consumers and producers, in comparison to copepods and protozoa.

Our objectives are:

1. Quantify feeding and growth rates of gonozooids at environmental and experimental conditions.

2. Quantify rates of asexual and longevity of phorozooid reproduction at environmental and experimental conditions
3. Quantify and compare feeding rates of gonozooids on a wide size range of food particles at environmental concentrations.

Although doliolids are abundant in numerous regions of the coastal ocean, and are considered to be major planktonic grazers, data on rates of feeding and growth are scarce. Chapter 1 of this thesis presents results on quantifications of grazing, ingestion, and growth rates of *D. gegenbauri* gonozooids in response to the ranges of food concentration and temperature as they may occur on the Southeastern continental shelf of the U.S.A. to understand at what rates doliolids function in their changing environment and how these rates may affect co-occurring planktonic organisms. Chapter 2 investigates phorozooid asexual production of gonozooids over their reproductive life at food concentrations and temperatures resembling their natural environment. Thus, determining if asexual reproduction can lead to rapid colonization of neritic regions. Chapter 3 revisits the grazing rates of *D. gegenbauri* gonozooids on large particles. It is hypothesized that the presence of large diatoms would affect the feeding of gonozooids of *D. gegenbauri* on smaller, co-occurring cells.

The goal of the present study is to determine clearance and ingestion rates of gonozooids from 3.5 to 5.5 mm length, with increasing abundances of *Rhizosolina alata*, a diatom predominant on the southeastern continental shelf throughout the year, and compare these data to previous experiments.

REFERENCES

Berner, L.D. 1967. Distributional atlas of Thaliacea in the California current region. Cal. Coop. Oc. Fish. Invest. Atlas No. 8: 1-322.

Binet, D. 1976. Contribution a l'écologie de quelques taxons du zooplankton de cote d' Ivoire. II. Dolioles- Salpes- Appendiculaires. Doc. Scient. Centre Rech. Océanogr. Abidjan 7, 45-61.

Braconnot, J.C. 1963. Etude du cycle annuel des Salpes et Dolioles en rade de Villefranche-sur-Mer. J. Cons. perm. int. Explor. Mer. 28 (1): 21-36:

Braconnot, J.S. 1971. Contribution à létude biologique et écologique des tuniciers pélagiques salpides et doliolides. I. Hydrologie et écologie des salpides. Vie et Milieu 22, 257-286.

Burkill, P.H., E.S. Edwards, A.W.G. John and M.A. Sleigh. 1993. Microzooplankton and their herbivorous activity in the northeastern Atlantic Ocean. Deep-Sea Res. II 40: 479-493.

Crocker, K.M., A.L. Alldredge and D.K. Steinberg. 1991. Feeding rates of the doliolid, *Dolioletta gegenbauri*, on diatoms and bacteria. J. Plankton Res. 13: 77-82.

Dam, H.G. C.A. Miller and S.H. Jonasdottir. 1993. The trophic role of mesozooplankton at 47EN, 20EW during the North Atlantic Bloom Experiment. Deep-Sea Res. 40: 197-212.

De Decker, A. 1973. Agulhas Bank plankton *In:* The Biology of the Indian Ocean, B. Zeitzschel, ed., Springer-Verlag, New York, pp. 189-220.

Deibel, D. 1982a. Laboratory determined mortality, fecundity and growth rates of *Thalia democratica* FORSKAL and *Dolioletta gegenbauri* ULJANIN. J. Plankton Res. 4: 143-153.

Deibel, D. 1982b. Laboratory-measured grazing and ingestion rates of the salp, *Thalia democratica* Forskal, and the doliolid, *Dolioletta gegenbauri* Uljanin (Tunicata,Thaliacea). J. Plankton Res. 4: 189-201.

Deibel, D. 1985a. Blooms of the pelagic tunicate, *Dolioletta gegenbauri*: Are they associated with Gulf Stream frontal eddies? J. Mar. Res. 43: 211-236.

Deibel, D. 1985b. Clearance rates of the salp *Thalia democratica* fed naturally occurring particles. Mar. Biol. 86: 47-54.

Deibel, D. 1995. Ecological impacts of pelagic tunicates. *In*: Q. Bone. ed. The Biology of Pelagic Tunicates, Oxford University Press (in press).

Esnal, G.B. and L.C. Simone. 1982. Doliolidos (Tunicata, Thaliacea) del Golfo de Mexico y Mar Caribe. Physis (Buenos Aires), Secc. A, 40: 51-57.

Godeaux, J. 1962. Tuniciers pélagiques. Inst. Royal Sci. Nat. de Belgique, Res. Sci. 3: 1 -32.

Harbison, G.R. and R.W. Gilmer. 1976. The feeding rates of th epelagic tunicate *Pegea confederata* and two other salps. Limnol. Oceanogr. 21: 517-528

Heron, A.C. 1972. Population ecology of a colonizing species: The pelagic tunicate *Thalia democratica*. I. Individual growth rate and generation time. Oecologia. 10: 269-293.

Heron, A.C. and E.E. Benham. 1984. Individual growth rates of salps in three populations. J. Plankton Res. 6:811-828.

Hopkins, T.L. and J.J. Torres. 1989. Midwater food web in the vicinity of a marginal ice zone in the western Weddell Sea. Deep-Sea Res. 36: 543-560.

Lindley, J.A., J. Roskell, A.J. Warner, N.C. Halliday, H.G. Hunt, A.W.G. John and T.D. Jonas. 1991. Doliolids in the German Bight in 1989: evidence for exceptional inflow into the North Sea. J. Mar. Biol. Ass. U.K. 70: 679-682.

Mackas, D.L., L. Washburn and S.L. Smith. 1991. Zooplankton community pattern associated with a California current cold filament. J. Geophys. Res. 96, No. C8 14781-14797.

Miller, C.B. 1993. Pelagic production processes in the Subarctic Pacific. Prog. Oceanog. 32:1-15.

Mullin, M.M. 1983. In situ measurement of filtering rates of the salp, *Thalia democratica*, on phytoplankton and bacteria. J. Palnkton Res. 5: 279-288.

Paffenhöfer, G.-A. 1976. On the biology of appendicularia of the southeastern North Sea. In: 10th Europ. Symp. Mar. Biol., Ostend, Belgium, 1975, Vol. 2, 437-455, G. Persoone and E. Jaspers, eds., Universa Press, Wetteren.

Paffenhöfer, G.-A. and T.N. Lee. 1987a. Development and persistence of patches of thaliacea. South Afr. J. Mar. Sci. 5: 305-318.

Paffenhöfer, G.-A. and T.N. Lee. 1987b. Summertime Gulf Stream intrusions into southeast U.S. shelf waters in 1981: Changes in distribution and abundance of particulate matter. Prog. Oceanog. 19: 373-401.

Paffenhöfer, G.-A., B.T. Wester and W.D. Nicholas. 1984. Zooplankton abundance in relation to state and type of intrusions onto the southeastern United Sates shelf during summer. J. Mar. Res. 42: 995-1017.

Paffenhöfer, G.-A., B.K. Sherman and T.N. Lee. 1987. Summer upwelling on the southeastern continental shelf of the U.S.A. during 1981: Abundance, distribution and patch formation of zooplankton. Prog. Oceanog. 19: 403-436.

Paffenhöfer, G.-A., L.P. Atkinson, T.N. Lee, P.G. Verity and L.R. Bulluck, III. 1995. Distributions and abundance of thaliaceans and copepods off the southeastern U.S. during winter. Cont. Shelf Res. 15: 255-280.

Seguin, G. and F. Ibanez. 1974. Zooplankton provenant de radiales effectuées le long de la côte sénégalaise par le navire océanographique Laurent Amaro en mai 1968. Bull. de l'inst. Found. d'Afrique Noire. Ser. A., Sci. Nat. 36: 842-879.

Sheard, K. 1965. Species groups in the zooplankton of eastern Australian slope waters, 1938-41. Aust. J. Mar. Fresh W. Res. 16: 219-254.

Tebeau, C.M. an L.P. Madin. 1994. Grazing rates for three life history stages of the doliolid *Dolioletta gegenbauri* Ulijauin (Tunicate, Thaliacea) J. Plankton Res. 16: 1075-1081.

Timonin, A.G. M.V. Flint and L.A. Zadorina. 1979. Seston (total plankton biomass) distribution in the coastal water off Peru. Oceanology 19: 200-204. (USSR, Engl. Transl).

CHAPTER II

FEEDING AND GROWTH RATES OF THE DOLIOLID,
DOLIOLETTA GEGENBAURI ULJANIN
(TUNICATA, THALIACEA)1

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[1] Gibson, D. M. and G.-A. Paffenhofer. 2000. Journal of Plankton Research. 22:1485-1500.

ABSTRACT

The goal of this research is to enhance our knowledge of the contributions of doliolids to the planktonic community as consumers and secondary producers. The objectives are to quantify feeding and growth rates of *Dolioletta gegenbauri* gonozooids at 4 food concentrations and 4 temperatures in order to determine their impact as grazers throughout the water column. Although doliolids are abundant in numerous regions of the coastal ocean, and are considered to be major planktonic grazers, data on rates of feeding and growth are scarce. Laboratory experiments were conducted at 16.5, 20, 23.5, and 26.5°C quantifying removal of a 50:50 volumetric concentration of *Thalassiosira weissflogii* and *Rhodomonas* sp. at four different food concentrations of 20, 60, 160, and 390 µg C · l^{-1}. Results from these experiments suggest that clearance rates are similar at concentrations from 20 to 60 µg C · l^{-1}, and decrease as the food concentrations increase to 160, and 390 µg C · l^{-1}. The ingestion rates increase over a range of phytoplankton concentrations from 20 to 160 µg C · l^{-1}, then decrease when abnormally high concentrations of 390 µg C · l^{-1} are offered. Clearance and ingestion rates increase as temperature increases from 16.5 to 26.5° C. The exponential growth rates range from k = 0.2 - 0.7 with the lowest rates occurring at the highest food concentration. Growth rates increase with increasing temperature from K = 0.1-0.3/day at 16.5° C to 0.45-0.7 at 26.5° C. In each case, the small and medium sized zooids had higher growth rates than the larger gonozooids. These results suggest that doliolid feeding and growth rates are a function of environmental food concentrations and temperatures, and imply that they can be important consumers in a changing neritic environment.

INTRODUCTION

During the most recent major zooplankton symposium (Harris, 1995) the emphasis was on production with the majority of presentations on protozoa and copepods. Earlier and more recent observations revealed high growth and production rates of pelagic tunicates (e.g. Heron 1972, Paffenhöfer 1976a, Hopcroft and Roff 1995), and also high abundances (e.g. Paffenhöfer 1976a, Deibel 1985). Field observations revealed that grazing by doliolids appeared to control the abundance of protozoa and free-spawning calanoid copepods (Paffenhöfer et al. 1995). Doliolids occur abundantly in neritic regions of subtropical oceans (e.g. Paffenhöfer and Gibson 1999, their Table I). To understand production rates, knowledge of rates of consumption is needed, particularly in relation to concentration and composition of food, size of the feeder and the range of environmental temperatures.

Grazing rate studies on doliolids are too few to generalize the response to changing conditions. There have only been four modern studies of feeding by doliolids (Madin and Deibel 1998). Deibel (1982b), reported on the laboratory-measured grazing and ingestion rates of *Thalia democratica* and *D. gegenbauri* over a range of food concentrations of 0.15-0.70 $mm^3 \cdot l^{-1}$ at 20° C, and on growth and reproduction rates in relation to controlled but environmentally realistic conditions. Deibel and Paffenhöfer (1988) developed methods for collecting and handling living doliolids which allowed them to observe the feeding mechanisms of *Doliolum nationalis* using high speed cinematography. Crocker *et al* (1991) studied the response of three life history stages of *D. gegenbauri* to naturally occurring food in two size ranges in order to more accurately assess the significance of these grazers in the planktonic food web. The purpose of Tebeau and Madin 1994 study was to quantify the grazing of trophozooids, phorozooids, and gonozooids of *D. gegenbauri* both in situ and under laboratory conditions closely resembling natural conditions. However, there have been no studies of the functional responses to food concentration, size, and quality, (Deibel, 1998), or to changes in feeding as a function of temperature.

The purpose of this study was to quantify grazing, ingestion, and growth rates of *D. gegenbauri* gonozooids in response to the ranges of food concentration and temperature as they may occur on the southeastern continental shelf of the U.S.A., and to understand at what rates doliolids function in their changing environment and how these rates may affect co-occurring planktonic organisms.

METHODS

Dolioletta gegenbauri oozooids, phorozooids, and gonozooids were collected throughout the year from the mid shelf waters along the Wassaw transect off the coast of Savannah, GA, onboard the RV Blue Fin. With the ship speed of $\leq$ 0.5 kt, a cone net of 0.5 m diameter, and 200 µm mesh with a 4 L codend was used to collect undamaged doliolids. Onboard ship the contents of the codend were gently floated into 5 gallon buckets, and up to 20 small undamaged, healthy zooids were separated into 2-L glass jars with a combination of Niskin bottle-collected seawater, and *Rhodomonas* sp., to ensure survival on the trip back to the Skidaway laboratory. The 2-L jars were then placed on a plankton wheel rotating at 0.2 rpm. to keep the phytoplankton and zooids in suspension in a light-dark cycle of 12h :12h, at 20°C. The zooids were allowed to acclimate to this environment for several days. Each day, at least 10% of the water along with aggregated particles, and fecal pellets were removed, and fresh unfiltered Niskin collected seawater was added.

D. gegenbauri were reared through several generations at 20° C, a temperature they are frequently encountered on the southeastern shelf (Deibel 1998), and at an average food concentration of 60 µg C · l $^{-1}$, equivalent to ~0.3 to >2mm ^{3} · l $^{-1}$ of particulate matter encountered offshore (Paffenhöfer and Lee 1987). A combination of food was offered, including the diatom *Thalassiosira weissflogii,* (~ 700 to 1,200µm ^{3}, 80 µg C · mm $^{-3}$), and the flagellate *Rhodomonas* sp., (~300 µm ^{3} volume, 160 µg C · mm $^{-3}$).

CHN analyses were conducted with laboratory reared gonozooids offered on average 60 µg C/l of a combination of *Rhodomonas* sp., and *Thalassiosira weissflogii* at 20° C to obtain a length to weight relationship (Fig. 1). The zooids were placed in 2-L jars with filtered seawater and allowed to evacuate their guts. The Sony - COHU video system for SZH microscopes was used to measure the length of live gonozooids, and 2 - 22 similar sized zooids were pooled onto ashed glass fiber filters to ensure a CHN reading higher than the blanks. The samples and blank filters were freeze dried for 24 hours and folded inside of ashed aluminum foil cups. Samples were then analyzed using a Fisons NA 1500. Each point on the graph represents 22 small to 2 large gonozooids from 2.5 to 8.5 mm length. These analyses enabled us to obtain enough data to construct a length vs. carbon content relationship. The power regression in Fig. 1 allowed for estimation of gonozooid carbon content of small to large gonozooids.

Feeding and growth experiments were conducted simultaneously with laboratory reared gonozooids. The feeding experiments followed two themes. First, several series of experiments was conducted at 20° C and four food concentrations of 20, 60, 120, and 390 µg C · l $^{-1}$. The second series of experiments was conducted at 60 µg C · l $^{-1}$ and four temperatures of 16.5, 20, 23.5, and 26.5º C. Each experiment was designed so that the mean experimental food concentration of *Rhodomonas* sp., and *Thalassiosira weissflogii* was the geometric mean of the initial and final concentration. This was achieved by selecting the correct number of zooids after calculating the estimated clearance rate obtained from initial work done by G. A- Paffenhöfer. Knowing the clearance rates of each gonozooid at a particular size, assured that the food concentration would not be reduced below 30%. After calculating the number of zooids to use, length measurements were made at the beginning and end of each experiment using the Sony video system. Almost all of the experiments were started with 2-3mm zooids. The same cohort was followed to obtain clearance and growth rates for the same animals to maturity of 6-7mm egg and sperm producing gonozooids. Clearance rates were measured by the differences between food concentrations at the beginning and end of the experiment using a Coulter Counter TAII. Phytoplankton growth constants in the controls, and the gonozooid clearance (ml · zooid $^{-1}$ · day $^{-1}$), and ingestion (µg C · zooid $^{-1}$ · d $^{-1}$) were calculated using the equations of Frost (1972) and Harbison and Gilmer (1976). Accuracy of the Coulter Counter was determined by analyzing slides prepared using an imaging cytometry system described by Verity and Sieracki (1993). Epi-fluorescence slides were made from a 50 ml sample from each control and experiment, along with the coulter samples, to compare the cells/ml counted. On average, the Coulter counts were 5% higher than the epi-fluorescence counts.

RESULTS

Concentration Dependent Clearance Rates

We offered identical volumetric abundances of *Rhodomonas* sp. and *T. weissflogii* at the start of each experiment. The concentrations chosen for our experiments, span the range of algal concentrations found on the southeastern continental shelf over the various seasons. The lower two concentrations are often found, and 160 µg C · l $^{-1}$ is regularly encountered in mature intrusions of cold Gulf Stream water. On very rare occasions is 390 µg C · l $^{-1}$ found in such upwellings (e.g. Yoder et al. 1985). Each point on the graph represents from 4-11 feeding experiments with gonozooids of *D. gegenbauri*. The Kruskal-Wallis test and the Multiple Comparisons test were used to determine the similarities and differences of clearance rates between food concentrations with an $\alpha = 0.05$. We chose zooids of three different weights to represent small, medium and large animals.

Clearance rates increased with increasing body weight. From 20 to 60 µg C · l $^{-1}$, the 5 and 15 µg C zooids had similar clearance rates, but the rates decreased significantly as concentration increased to 390µg C · l $^{-1}$ (Fig. 2). The large zooids of 35 µg C had similar clearance rates at 20, 60 and 160 µg C · l $^{-1}$, then significantly decreased at the highest concentration.

Concentration Dependent Ingestion Rates

Ingestion rates increased with increasing food concentration from 20 to 160 µg C · l $^{-1}$ and then decreased at 390 µg C · l $^{-1}$ (Fig. 3). Ingestion rates for the small gonozooids did not change significantly over the four food concentrations. However, clearance rates for the medium and large zooid increased significantly from 20 to 160 µg C · l $^{-1}$, and then decreased at the highest concentration. Weight-specific ingestion rates ranged from 60 to 156% for 5 µg, from 52 to 173% for 15 µg, and from 30 to 198% for 35 µg zooids. Maximum rates were found at 160 µg C · l $^{-1}$, increasing with increasing body weight. Minimum rates at 20 µg C · l $^{-1}$ decreased with increasing body weight.

Concentration Dependent Growth Rates

Daily growth rates for 15 µg C zooids increased with increasing body weight over food concentrations of 20 to 60 µg C · l^{-1}, did not change at 160, and decreased towards 390 µg C · l^{-1} (Fig. 4). Growth rates for 5 and 35 µg C zooids showed no significant differences for the four different food levels. Gonozooids at 5 and 15 µg C had higher growth rates than the largest zooids. Maximum rates were attained by the 15 µg C zooids at 60 and 160 µg C · l^{-1}.

Temperature Dependent Clearance Rates

We chose a range of temperatures at which *D. gegenbauri* had been found abundantly on the southeastern continental shelf (Deibel 1985, Paffenhöfer et al. 1991), and a food concentration of 60 µg C · l^{-1} (~1.5 µg chlorophyll/l), near which doliolids occurred abundantly (Paffenhöfer et al. 1995). Each point represents from 4 to 12 experiments (Fig. 5). Clearance rates of small zooid did not change significantly with increasing temperature. Medium-sized zooids rates increased significantly from 16.5º to 20º, remained similar at 23.5º, and significantly increased at 26.5º (Fig. 5). The largest zooids' clearance rates significantly increased from 16.5 to 26.5ºC, attaining near 1,400 ml swept clear · zooid $^{-1}$ · d $^{-1}$.

Temperature Dependent Ingestion Rates

Ingestion rates followed the same trend as the clearance rates, where the smaller zooids had similar rates over the temperature range at, 4.6, 6.5, 7.0, and 6.0 µg C · zooid $^{-1}$ d $^{-1}$. They ingested between 92 to 140% of their body carbon daily. The 15 µg C zooids ingested from 59 to 147 %, and the large zooids from 43 to 179% of their body carbon every day (Fig. 9). The small zooid hardly changes its rate with changing temperature whereas the large zooid has the strongest increase in ingestion rate with increasing temperature (Fig. 6).

Temperature Dependent Growth Rates

The growth rates for 5 µg C zooid increased with increasing temperature from k = 0.30 to 0.60 (Fig. 7). Growth rates for the 15 and 35 µg C zooids both decreased from 20º to 23.5º C despite a sufficient number of experiments ranging from 5 to 12. The rather low rates for 15 and 35 µg g C zooids at 23.5º C are followed by increases at 26.5º C. The small and medium zooids had similar growth rates as compared to the growth rates for the large zooids which were the lowest, except at 20º C.

DISCUSSION

General Comments

When discussing our results, we will refer to information from studies of doliolids, salps, appendicularia and copepods. Doliolids obtain food via motion of cilia, whereas salps have to swim to force water to enter their mouths; appendicularia move their tail rhythmically to propel water into their house, while many calanoid copepods species (copepodid stages and adults) produce a feeding current (e.g. *Eucalanus pileatus, Temora* spp.), allowing them to perceive individual cells at a distance. The genus *Acartia* to which we will refer repeatedly, does not produce such a current and uses different means to capture food (e.g. Paffenhöfer and Stearns 1988).

The food species offered do not encompass the wide range of abundant natural particles encountered on the southeastern shelf by doliolids. While small diatoms can be found at up to 1.5 mm^3/l of cell volume, maximum abundances of small flagellates are considerably lower (Paffenhöfer, pers. obs.). In order to have small and large zooids ingest the same food species, we chose *Rhodoosemonas* sp. and *T. weissflogii*.

Weight versus Length Relationship

We established weight-length relations to be able to run experiments without sacrificing zooids every day for body carbon quantifications. However, we were only able to establish this relationship for average food concentrations of *Rhodomonas* sp. and *T. weissflogii* near 60 µg C $\cdot$ l $^{-1}$ and 20° C. Results for *Acartia hudsonica* indicate that the body weight/carbon relationship varies directly as a function of food concentration (Durbin and Durbin, 1992). Over a range of food concentrations from 8 to 590 µg C/l, the increases in weight of adult female *A. hudsonica* ranged from less than 5% to near 28% of the body carbon at the lowest food level (Fig. 5 of Durbin and Durbin 1992) with averages near 15%. Our range was from 20 to 390µg C $\cdot$ l $^{-1}$. We decided not to make corrections for the effects of changing food concentrations or temperatures on the size to body carbon relationship. One equation was used for all temperatures and food concentrations, assuming that the error in weight/carbon content would not surpass 15% of the values obtained at 60 µg C $\cdot$ l $^{-1}$ and 20° C, similar to Deibel (1982a,b).

Clearance Rates

We used gonozooids that had been reared in the laboratory, i.e. were not collected in the ocean, and therefore were undamaged for our experiments. There are no earlier data relating clearance rates over a range of food concentrations for doliolids, except Deibel (1982b) who quantified weight-specific clearance rates for gono- and oozooids vs. initial food concentration. Clearance rates of copepods increase with decreasing food concentration for virtually all species studied, eventually attaining a maximum (at ~ 25µg C $\cdot$ l ^{B1} for *Acartia hudsonica*, Durbin and Durbin 1992; and *A. tonsa* Paffenhöfer and Stearns 1988; near 8 µg C $\cdot$ l ^{B1} for *Eucalanus pileatus*, Paffenhöfer and Lewis 1990; and also *E. hyalinus* Paffenhöfer 1988; each of these species feeding on a different type of phytoplankton food!), and then decreasing or remaining constant as food levels decrease further. In our studies, the clearance rates of *D. gegenbauri* gonozooids increased with decreasing food concentration from 390 to 60 µg C · l $^{-1}$. Those rates remained constant or decreased slightly at 20 µg C · l $^{-1}$ (Fig. 2). E. *pileatus* can increase its clearance rate toward very low phytoplankton levels (< 10 µg C/l), whereas *D. gegenbauri* cannot. The ecological implications of that finding will be evaluated later.

As for clearance rates vs. temperature for metazooplanknton, results on copepods are the only data available for comparison (e.g. Deason 1980, Durbin and Durbin 1992). Maximum clearance rates for adult *Acartia hudsonica* females feeding on *Skeletonema costatum* increased from 5º to 10º, and decreased from 10 to 15º C (Deason 1980). This was accompanied by a decrease in copepod weight with increasing temperature. Durbin and Durbin, 1992 found the same trend for females of the same species as temperatures increase from 4.5º to 16º C.

Clearance rates of *Dolioletta gegenbauri* were determined not only at four different temperatures but also for three different gonozooid sizes (Fig. 5). The clearance rates of the largest zooid increased by a factor of 5 from 16.5º to 26.5º C, whereas the rates for the smallest zooid remained unchanged over the same temperature range. The medium-sized zooids increased their clearance rate by factor 2.5 from 16.5º to 26.5º C. However, early juvenile gonozooids appear to be unable to compensate increasing metabolic expenditures with increasing temperature by an increase in clearance rate (Fig. 5). In essence, temperature affects clearance rates positively with increasing size of the gonozooids.

Ingestion Rates

Ingestion rates of macrozooplankton, including juveniles, increase with increasing food concentration: e.g. Frost (1972) for the copepod *Calanus pacificus*, Durbin and Durbin (1992) for the copepod *Acartia hudsonica*, Paffenhöfer (1984) for juveniles of the copepod *Paracalanus* sp. Beyond a critical concentration, the ingestion rates of many of the investigated copepod species remain constant (e.g. Frost 1972, Durbin and Durbin 1992). One notable exception were females of *Calanus hyperboreus* whose ingestion rate increased to a maximum and then decreased with increasing phytoplankton concentration (Mullin 1963). Our observations on three different sizes of gonozooids resemble Mullin′s observation, i.e. ingestion rates increase from 20 to 160 µg $C \cdot l^{-1}$ (Fig. 3). It appears as that the high phytoplankton concentration affects the feeding behavior of gonozooids of all sizes studied.

Weight-specific ingestion rates also show the abovementioned increase and decrease (Fig. 8). These rates, are far higher for the small than for the large zooid at the two lower food concentrations. However, the large zooid's ingestion rate exceeds the small and medium-sized zooids at the two higher food levels. One explanation could be that the mouth opening of the small zooid allows a higher water flow rate vs. body weight than the larger zooid. At $\geq$160 µg $C \cdot l^{-1}$ the relative advantage of higher flow rate per unit body weight could be overcome by the ability of the large zooid to process food. A comparison of these rates with those of calanoid copepods at similar temperatures reveals similarities at concentrations ranging from ~ 20 to ~ 160 µg $C \cdot l^{B1}$ (Table 1).

The temperature dependent ingestion rates (Fig. 6) are a mirror-image of the temperature related clearance rates (Fig. 5), indicating an eventual decrease with increasing temperature for the small zooid, and increases for the two larger sizes. Weight specific ingestion rates of the small gonozooid changed slightly as the temperature increased when compared to the two larger size gonozooids (Fig. 9). The smaller gonozooids were capable of ingesting a constant amount of carbon per body weight as the temperature increased, where as the two larger size gonozooids were able to significantly increase their carbon intake to compensate for the increased metabolic expenderatures at high temperatures. At a lower temperature range, maximum weight specific ingestion rates of adult females of *Acartia hudsonica* showed little change from 4.5º to 8º C (44 and 37%, respectively), and increased to 68% at 12º and 93% at 16º C (Durbin and Durbin 1992). *D. gegenbauri* of 35 µg C weight-specific ingestion rates increased from 43% at 16.5º to 176% at 26.5º C, which indicates that temperature has a positive affect on ingestion rates of doliolids and copepods. However the latter can ingest more carbon/body weight at the same temperature, 93% at 16º C, as compared to 43% for doliolids.

Growth Rates

Exponential growth rates were quantified from gonozooid assemblages growing from near 2mm to near 8mm zooid length, their length being measured daily, and several zooids removed daily to always have no more than a 30% decrease of the initial food concentration. Our data reveal limited changes in k (exponential growth rate) vs. food concentration for the small zooids (Fig. 4). The growth rates of both medium and large gonozooids increased from 20 to 60 µg C · l^{1}, remained the same at 160 µg C · l ^{B1} and then decreased toward 390 µg C · l ^{B1}. The large zooids had the lowest growth rates at each food concentrations probably due to a general decrease in somatic growth near the maximum size which may be attributed to the start of developing ovaries/testes, or to an overall decrease in gross growth efficiency. The unusually high growth rate of the small zooid at 20 µg C · l ^{B1} is supported by 11 independent experiments, and that at 60 µg C · l ^{B1} by 8. It may be possible that the condition of the usually well fed phorozooid (~ 50 to 100 µg C · l ^{B1}) improved growth of the gonozooid during the first two days after it separated from the phorozooid. The exponential growth rates ranged from 0.20 to 0.71 (Fig. 4). Deibel (1982a) reported growth rates of 0.03 to 0.36 for gonozooids, and of 0.17 to 0.69 for phorozooids of *D. gegenbauri* at 20º C at ~70 to 100 µg C · l ^{B1} of *Isochrysis galbana*. At similar temperatures the appendicularian *Oikopleura dioica* had a k > 1.0 (Fenaux 1977). The salp *Thalia democratica* with a length increase of 8% · h ^{B1} 19 to 23º C (Tsuda and Nemoto 1992) would have had a daily exponential growth rate of 4.2 using Deibel=s (1982a) length vs. weight regression. Heron and Benham (1984) reported even greater *in situ* hourly length increases for *T. democratica*. Maximum growth rates were highest for the early copepodid stages of *Calanus pacificus* at food concentrations from ~ 50 to ~ 670 µg C · l ^{B1} (Vidal 1980). Similar exponential growth rates results were obtained for nauplius III to young adult female *C. helgolandicus/pacificus* at 15ºC (Paffenhöfer 1976b). between 41 and 101 µg C · l ^{B1}, with a decreased from 0.29 to 0.13 at the lowest and from 0.40 to 0.20 at the highest food level form nauplius to early adult.

Doliolid growth rates were lower than those of appendicularia and salps at similar temperatures, were higher than calanoid copepods at similar stages. There is no obvious explanation as to why doliolids grow slower than the other two tunicate taxa at similar food levels.

The effect of temperature on growth rates of *D. gegenbauri* was greater and more variable for the medium and large zooids (Fig. 7). Rates at 20º C were higher than those at 23.5º C for these two zooid sizes; the number of experiments were 5 at 20º but significantly the same, and

12 at 23.5º for the medium, and 7 and 5 respectively, for the large zooid. Presently, we do not have an explanation for the unexpected decrease in k from 20º to 23.5º C for the medium and large zooids. Similarly, clearance and ingestion rates of the 15 µg C zooids were lower at 23.5º C than at 20º C (Figures 5 and 6). The large zooids had the lowest rates, as expected, whereas the other two zooid sizes had similar rates (Fig. 7). In comparison, daily growth rates of *Acartia hudsonica* copepodid stage I to V increased rather evenly from 8% at 4.5º to 33% at 16º C (Durbin and Durbin 1992). *A. hudsonica* favors this range of temperatures, and *C. pacificus* that from ~ 7 to 16º C. The latter=s growth rates increased for all stages from CII to V from 8º to 15º C (Vidal 1980). However, the difference between maximum growth rates for a particular stage decreased for older animals. The rates at 15.5º C surpassed those at 8º C by > 100% for CII at > 200 µg C · l ^{B1}, and the rates for CV at the three different temperatures were nearly identical. Since food concentrations were used in our temperature study were not satiating, only a cautious comparison is in order. It is unlikely that similar results could be found for *D. gegenbauri* as the largest zooid=s growth rate at 16.5EC was by far the lowest of all and five times lower than that at 26.5º C (Fig. 5). This low temperature is more unfavorable to the large gonozooids than to the smaller ones.

Feeding Performance at High vs. Low Food Levels

When evaluating feeding rates of *D. gegenbauri*, we noticed that clearance rates were essentially the same at food concentrations of 60 and 20 µg C · l ^{B1} (Fig. 2). This is quite different for copepods whose clearance rate increased at food concentrations as low as 8 µg C · l ^{B1} or even lower (e.g. Paffenhöfer 1988, Paffenhöfer and Lewis 1990). Therefore, we attempted to compare feeding rates of a copepod and a gonozooid of similar size to obtain some insight (Table 2). Gonozooid clearance rates remained constant with decreasing food levels (Fig. 2), whereas the copepod=s rate increased by a factor of 6. The doliolid=s weight specific ingestion rate decreased from about 210% of its body carbon per day to 32%/d over an 8-fold food level decrease, while the copepod=s weight specific ingestion rate only decreased from 43 to 27% over a 10-fold decrease in food concentration. Copepods appear to be able to increase their clearance rate as food levels decrease by elevating their food-perception performance (Paffenhöfer and Lewis 1990) whereas doliolids cannot. This may not affect doliolid populations at food levels $\geq$ 20 µg C · l ^{B1} (Figure 3 and 4). However, on the southeastern shelf of the U.S., concentrations of phyto- and protozooplankton carbon can be often below 10 µg C · l ^{B1}. If the doliolid=s clearance rate were near 510 ml · zooid $^{-1}$ · d $^{-1}$ at a food level of 8 µg C · l $^{-1}$, it would ingest only 14% of its body carbon per day. Two points could be made here: (1) *D. gegenbauri* gonozooids can take advantage of high food concentrations by ingesting high percentages of their body weight; however, (2) they cannot compensate for decreases in food concentration below 60 µg C · l ^{B1}by increasing their clearance rates, as many calanoid copepods can (e.g. Frost 1972, Paffenhöfer 1988).

The results of studies on asexual reproduction rates and gross growth efficiencies of *D. gegenbauri* at the various food levels will allow us to obtain a comprehensive picture on the effect of food concentration on doliolid propagation and persistence, as compared to that of co-occurring calanoid copepods.

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REFERENCES

Bone, Q. (ed.) (1998) *The Biology of Pelagic Tunicates*. Oxford University Press, Oxford, 340 pp.

Crocker, K. M., Alldredge, A.L., and Steinberg, D. K. (1991) Feeding rates of the doliolid, *Dolioletta gegenbauri*, on diatoms and bacteria. *J. Plankton Res*.: 13, 77 - 82.

Deason, E.E. (1980) Grazing of *Acartia hudsonica (A. clausi)* on *Skeletonema costatum* in Narragansett Bay (USA): Influence of food concentration and tempearture. Mar. Biol., 60: 101-113.

Deibel, D. (1982a) Laboratory determined mortality, fecundity and growth rates of *Thalia democratica* Forskal and *Dolioletta gegenbauri* Uljanin (Tunicata, Thaliacea). *J. Plankton Res*.: 4, 143 - 153.

Deibel, D. (1982b) Laboratory- measured grazing and ingestion rates of the salp, *Thalia democratica* Forskal and the doliolid, *Dolioletta gegenbauri* Uljanin (Tunicata, Thaliacea). *J. Plankton Res*.: 4: 189 - 201.

Deibel, D. (1985) Blooms of the pelagic tunicate, *Dolioletta gegenbauri*: Are they associated with Gulf Stream frontal eddies? J. Mar. Res. 43: 211-236.

Deibel, D., and Paffenhöfer, G, -A. (1988) Cinematographic analysis of the feeding mechanism of the pelagic tunicate *Doliolum nationalis*. *Bull. Mar. Sci.*, 43: 404-412.

Durbin, E.G. and A.G. Durbin. (1992) Effects of temperature and food abundance on grazing and short-term weight change in the marine copepod *Acartia hudsonica*. *Limnol. Oceanogr*. 27: 361-378.

Fenaux, R. (1997) Life history of appendicularia (Genus *Oikopleura*). In *Proceedings of the Symposium on Warm Water Zooplankton*, National Institute of Oceanography, Goa. Pp. 497-510.

Frost. B. W. (1972) The effects of size and concentration of food particles on the feeding behavior of the marine planktonic copepod *Calanus pacificus*. *Limnol. Oceanogr*.: 17(6): 805-815.

Harbison, G. R., and Gilmer, R., W. (1976) The feeding rates of the pelagic tunicate *Pegea confoederata* and two other salps. *Limnol. Oceanogr.*, 121: 517-528.

Heron, A.C. (1972) Population ecology of a colonizing species: The pelagic tunicate *Thalia democratica*. I. Individual growth rate and generation time. Oecologia 10: 269-293.

Heron, A.C. and E.E. Benham. (1984) Individual growth rates of salps in three populations. *J. Plankton Res*. 6: 811-828.

Hopcroft, R.R. and J. C. Roff. (1995) Zooplankton growth rates: extraordinary production by the larvacean *Oikopleura dioica* in tropical waters. *J. Plankton Res*. 17: 205-220.

Madin, L. P., and Deibel, D. (1998) Feeding and energetics of Thaliacea. In Bone, Q. (ed.) (1998) *The Biology of Pelagic Tunicates*. Oxford University Press, Oxford, 340 pp.

Mullin, M.M. (1963) Some factors affecting the feeding of marine copepods of the genus *Calanus*. *Limnol. Oceanogr*. 8: 239-250.

Paffenhöfer, G.-A. (1976a) On the biology of appendicularia of the Southeastern North Sea. IN: Proc. 10. Europ. Symp. Mar. Biol., Ostend, Belgium, Vol. 2, Population Dynamics, G. Persoone and E. Jaspers, eds. Universa Press, Wetheren, Belgium, pp. 437-455.

Paffenhöfer, G.-A. (1976b) Feeding, growth, and food conversion of the marine planktonic copepod *Calanus helgolandicus*. *Limnol. Oceanogr*. 21: 39-50.

Paffenhöfer, G.-A. (1984) Food ingestion by the marine plankton copepod *Paracalanus* in relation to abundance and size distribution of food. Mar. Biol. 80: 323-333.

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

Paffenhöfer, G.-A. and S.C. Knowles. (1978) Feeding of marine planktonic copepods on mixed phytoplankton. Mar. Biol 48: 143-152.

Paffenhöfer, G.-A. and T.N. Lee. (1987) Development and persistence of patches of Thaliacea. South. Afr. Mar. Sci. 5: 305-318.

Paffenhöfer, G.-A. and D.E. Stearns. (1988) Why is *Acartia tonsa* restricted to the nearshore environment? Mar. Ecol. Progr. Ser. 42: 33-38.

Paffenhöfer, G.-A. and K.D. Lewis (1990) Perceptive performance and feeding behavior of calanoid copepods. J. Plankton Res. 12: 933-946.

Paffenhöfer, G.-A. and D.M. Gibson. (1999) Determination of generation time and asexual fecundity of doliolids (Tunicata, Thaliacea). J. Plankton Res. 21: 1193-1189.

Paffenhöfer, G.-A., T.B. Stewart, M.J. Youngbluth and T.G. Bailey (1991). High-resolution vertical profiles of pelagic tunicates. J. Plankton Res. 13: 971-981.

Paffenhöfer, G.-A., L.P. Atkinson, T.N. Lee, P.G. Verity and C.R. Bulluck, III (1995). Distribution and abundance of thaliaceans and copepods off the southeastern U.S.A. during winter. Cont. Shelf Res. 15: 255-280.

Tebeau, C. M., and Madin, L. P. (1994) Grazing rates for three life history stages of the doliolid *Dolioletta gegenbauri* Uljanin (Tunicata, Thaliacea). *J. Plankton Res.*: 16: 1075-1081.

Tsuda, A. and Nemoto, T. (1992) Distribution and growth of salps in a Kuroshio warm-core ring during summer 1982. *Deep-Sea Res.* 39 (Suppl. 1), S219-S229.

Verity, P.G. and Siereachki, M.E. (1992) Use of color image analysis and epifluorescence microscopy to measure plankton biomass. In Kemp, P.F., Sherr, B.P., Sherr, E.B. and Cole, J.J. (eds), *Handbook of Methods in Aquatic Microbila Ecology*. Lewis Publ., London, pp. 327-338.

Vidal, J. (1980) Physioecology of zooplankton. I. Effects of phytoplankton concentration, temperature, and body size on the growth rate of *Calanus pacificus* and *Pseudocalanus* sp. *Mar. Biol.* 56, 111-134.

Yoder, J.A., Atlkinson, L.P., Bishop, S.S., Blanton, J.O., Lee, T.N. and Pietrafesa, L.J. Phytoplankton dynamics within Gulf Stream instrusions on the southeastern United States continental shelf during summer 1981. *Cont. Shelf Res.,* 4, 611-635.

Zooplankton Production. (1995) Proceedings of a Symposium, Plymouth, England, 15-19 August 1994, R. Harris, ed ICES J. mar. Sci. 52: 261-773.

Table 1. Weight-specific ingestion rates of various species and stages of calanoid copepods.

Species and Stage	Food Type and Concentration (μgC·l-1)	Temperature (ºC)	Weight-Specific Ingestion Rate (μgC·μgC-1·d-1)	Reference
Acartia hudsonica (female)	*Thalassiosia constricta*	16		Durbin and Durbin 1992
	250		0.90	
	400		0.92	
A. hudsonica (female)	*Skeletonema costatum*	15		Deason 1980
	20		0.27	
	60		0.80	
	160		2.13	
Eucalanus pileatus Copepodid	*Leptocylindrus danicus* 17	20	0.54	Paffenhöfer and Knowles 1978
stages	the above species and		0.82	
	S. costatum, Rhizosolenia sp., 151		1.31	
Temora turbinata early - late copepods	*S. costatum* + *L. danicus* + *R. alata* 141	20	decreased from 1.65 to 0.62 with incr. Copepodid size	
Paracalanus sp.	*Thalassiosira weissflogii*	20		Paffenhöfer 1984
	24		1.10 for early copepodids 0.68 for late copepodids	
	80		1.90 for early copepodids 1.35 for late copepodid	

Table 2. Comparison of ingestion rates of *Eucalanus pileatus* and *Dolioletta gegenbauri* at 20°C at high and low phytoplankton concentrations. *E. pileatus* was feeding on *T. weissflogii*, and *D. gegenbauri* on a combination of *Rhodomonas* sp. and *T. weissflogii.*

	Food Concentration (µg C / l $^{-1}$)			
	8	20	80	160
E. pileatus (23 µg C body weight)				
Adult female				
Clearance (ml/copepod $^{-1}$ / d $^{-1}$)	506		82	
Ingestion (µg C/copepod $^{-1}$ / d $^{-1}$)	6.2		10	
Weight-specific ingestion (µg C/ µg C $^{-1}$ / d $^{-1}$)	0.27		0.43	
D. gegenbauri (35 µg C body weight)				
Mature gonozooid				
Clearance (ml /zooid $^{-1}$ /d $^{-1}$)		510		524
Ingestion (µg C/zooid $^{-1}$ /d $^{-1}$)		10.6		69.4
Weight specific ingestion (µg C/µg C $^{-1}$ /d $^{-1}$)		0.32		2.10

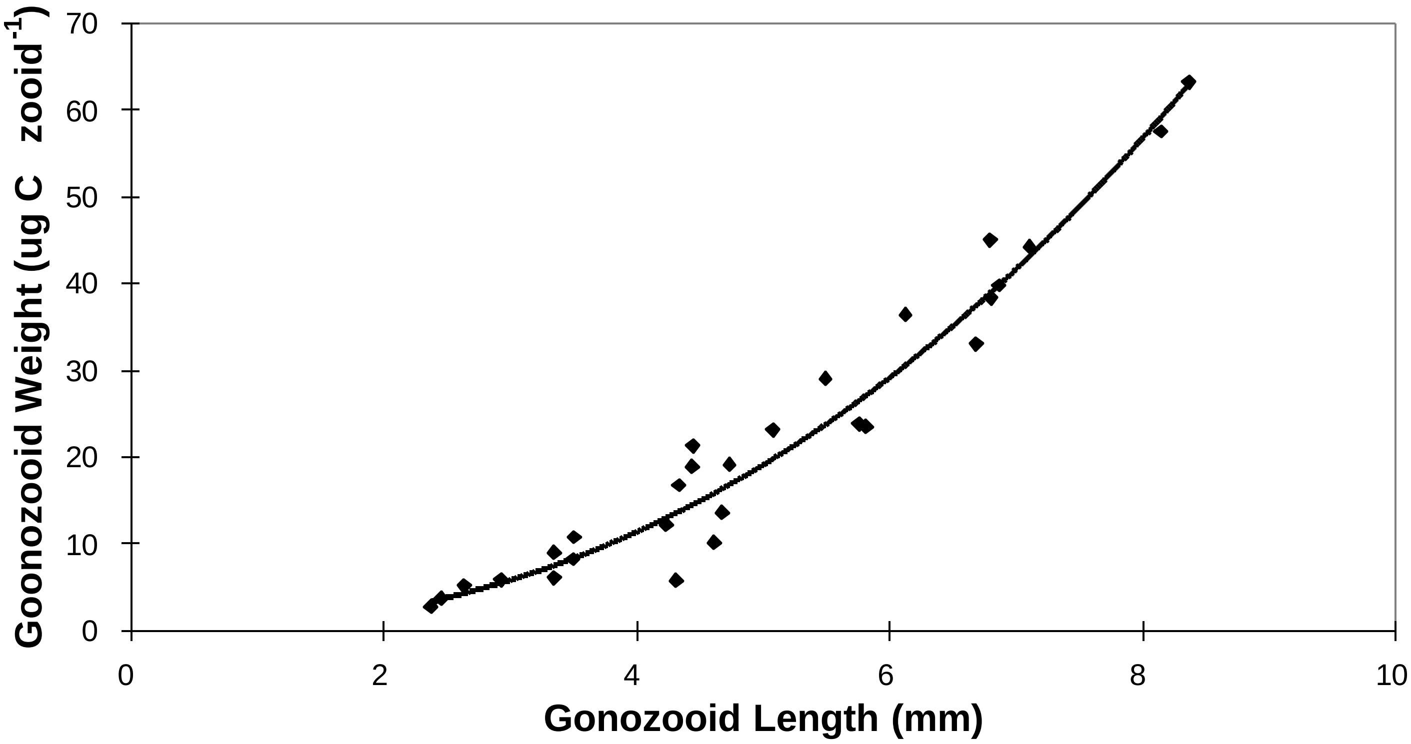

Figure 1. Weight (µg C) versus length (mm) for *Dolioletta gegenbauri* gonozooids. Each point is the result of pooling 2 - 22 gonozooids. The best-fit power function equation is w(µg C) = $0.4643x^{2.3119}$, n = 29, R^2 = .919.

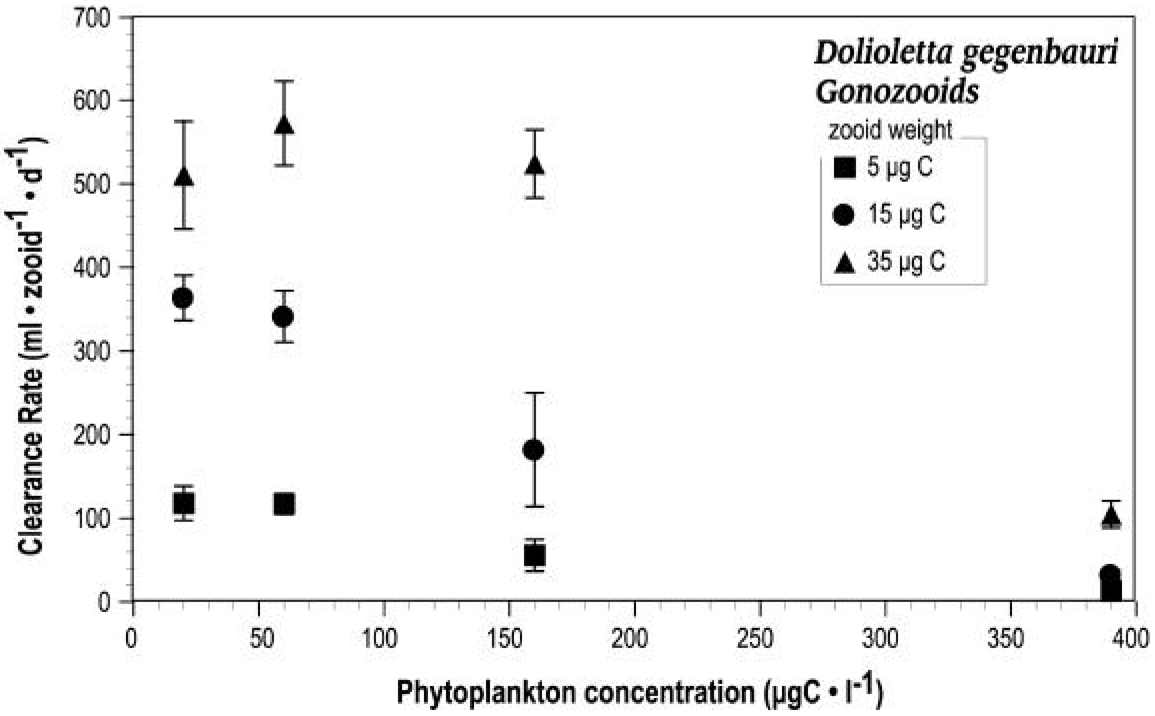

Figure 2. Mean (± SE) clearance rates (ml • zooid^{-1} • d^{-1}) versus phytoplankton concentration (µg C • l^{-1}) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 4 - 11 observations.

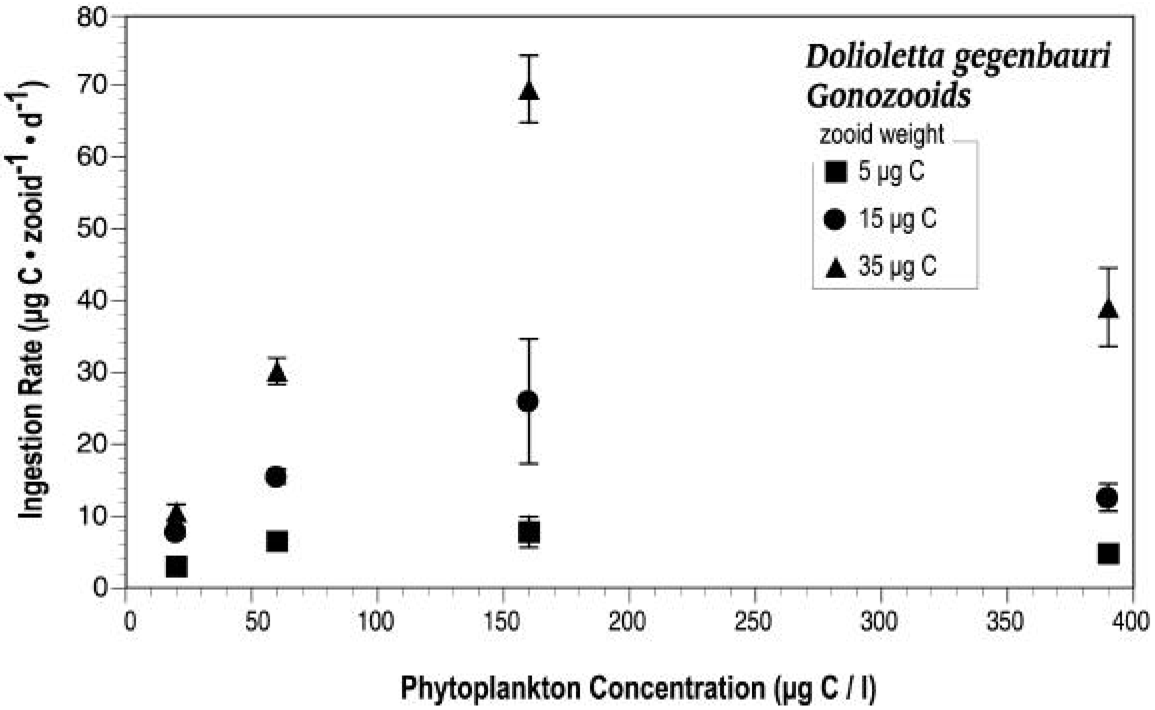

Figure 3. Mean (± SE) ingestion rates (µg C •zooid $^{-1}$ •d $^{-1}$) versus phytoplankton concentration (µg C • l $^{-1}$) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 4 - 11 observations.

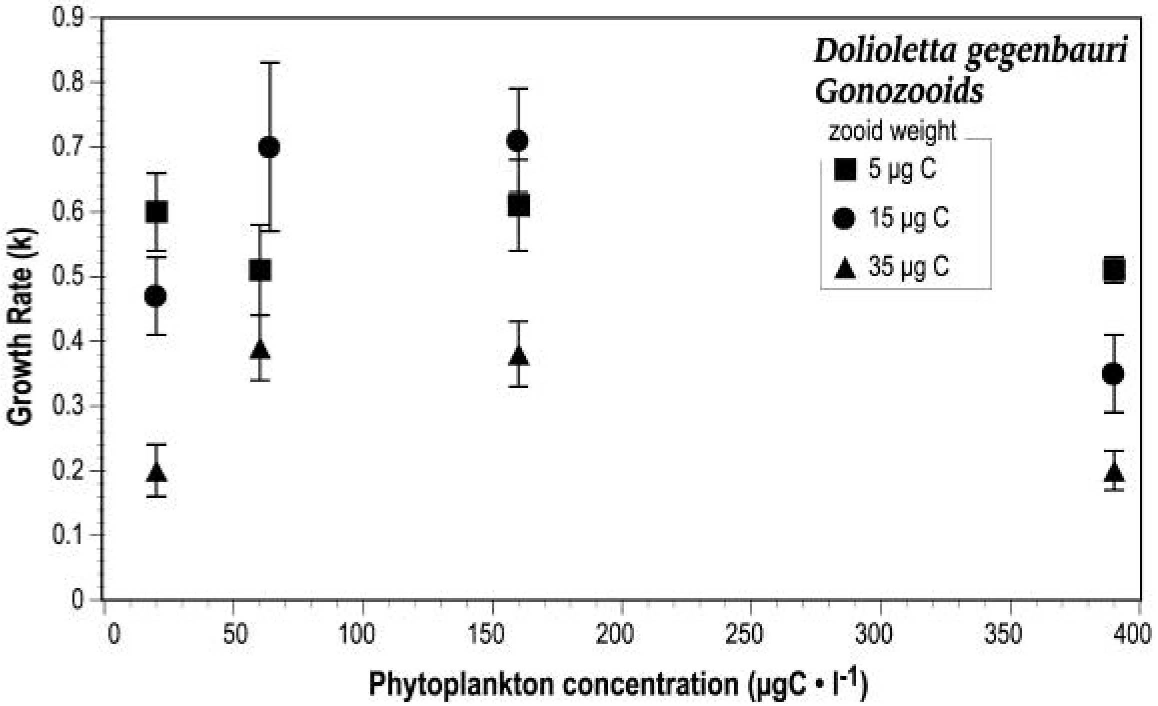

Figure 4. Mean (± SE) growth rates (*k*) versus phytoplankton concentration (µg C • l^{-1}) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 4 - 11 observations.

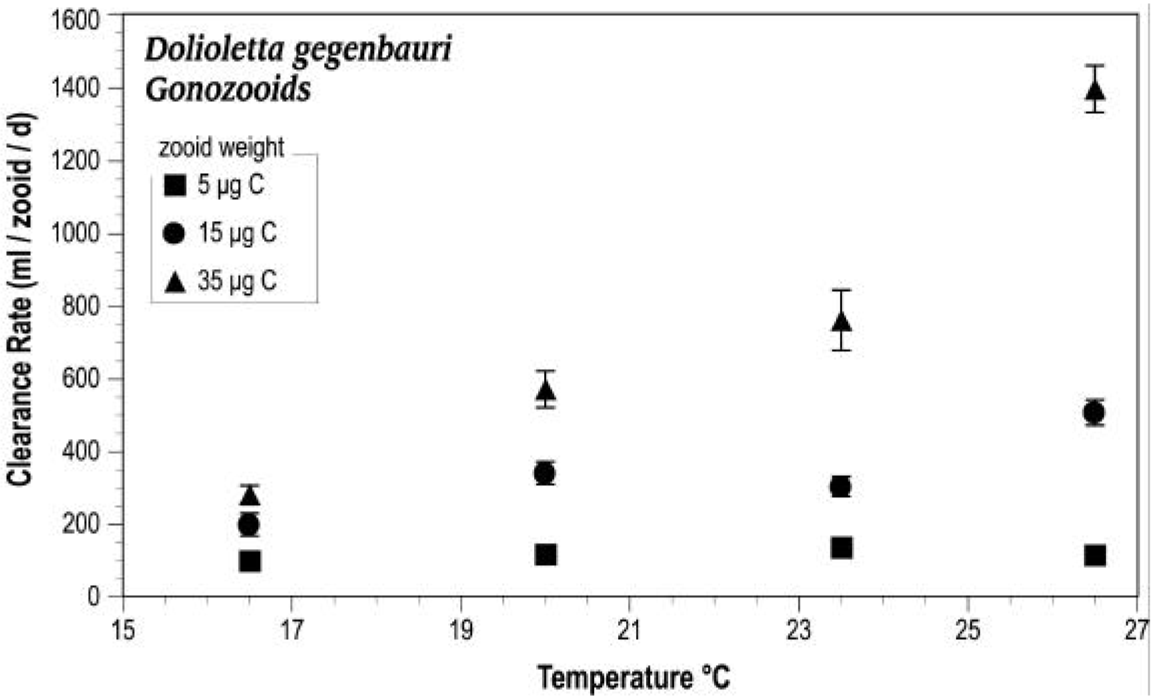

Figure 5. Mean (± SE) clearance rates (ml • zooid^{-1} • d^{-1}) versus temperature (^{o}C) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 4 - 12 observations.

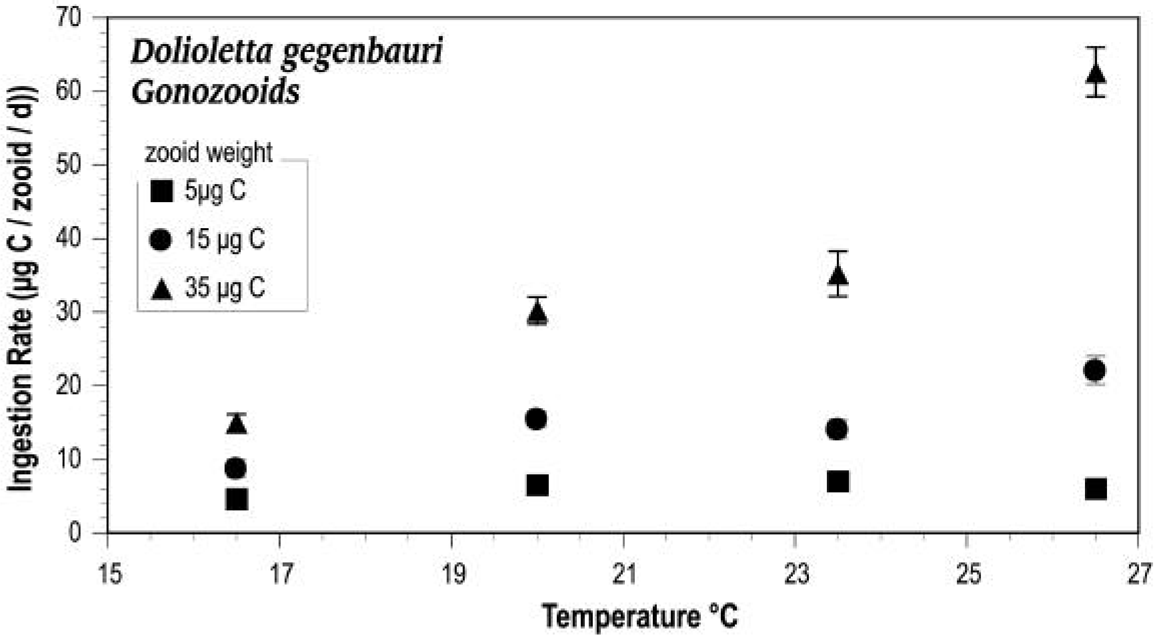

Figure 6. Mean (± SE) ingestion rates (µg C •zooid^{-1} • d^{-1}) versus temperature (^{o}C) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 4 - 12 observations.

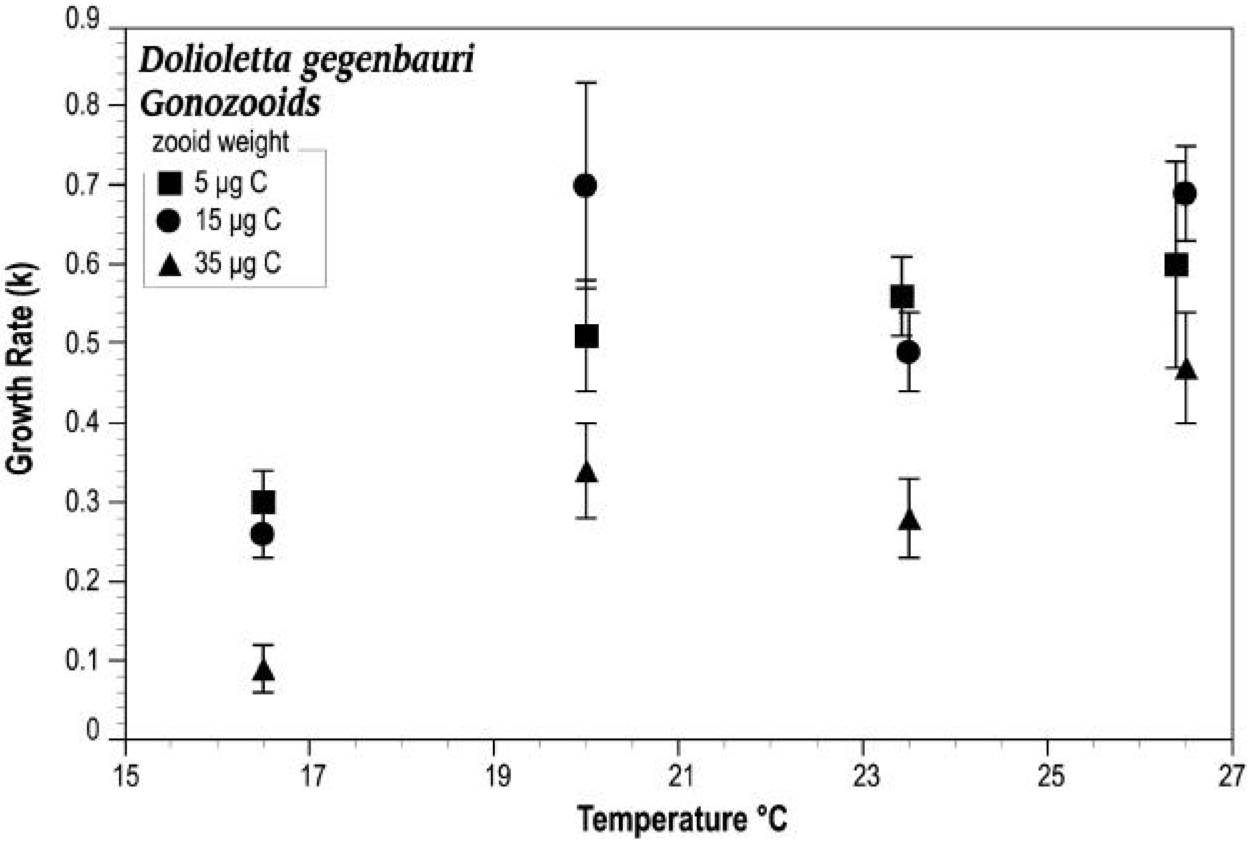

Figure 7. Mean (± SE) growth rates (*k*) versus temperature (°C) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 4 - 12 observations.

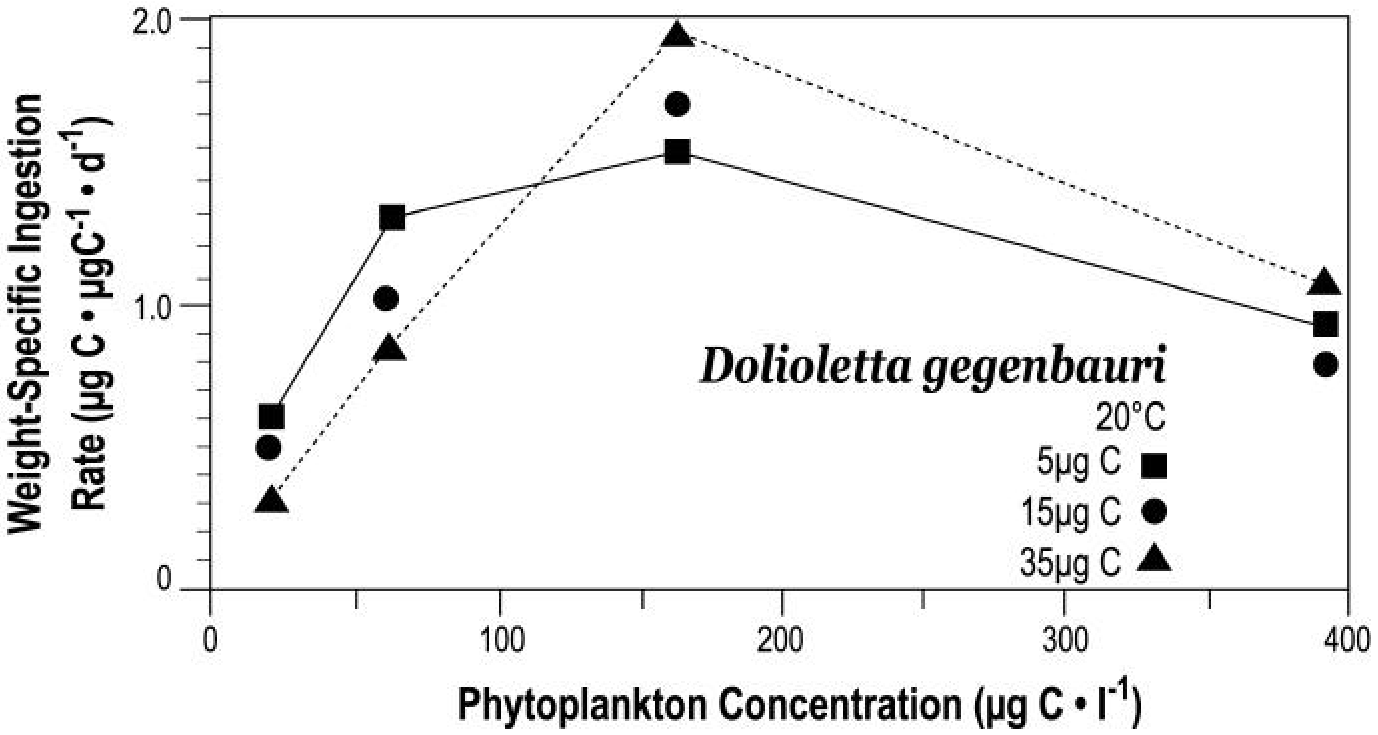

Figure 8. *Dolioletta gegenbauri*. Weight-specific ingestion rates of gonozooids of three different sizes in relation to phytoplankton concentration.

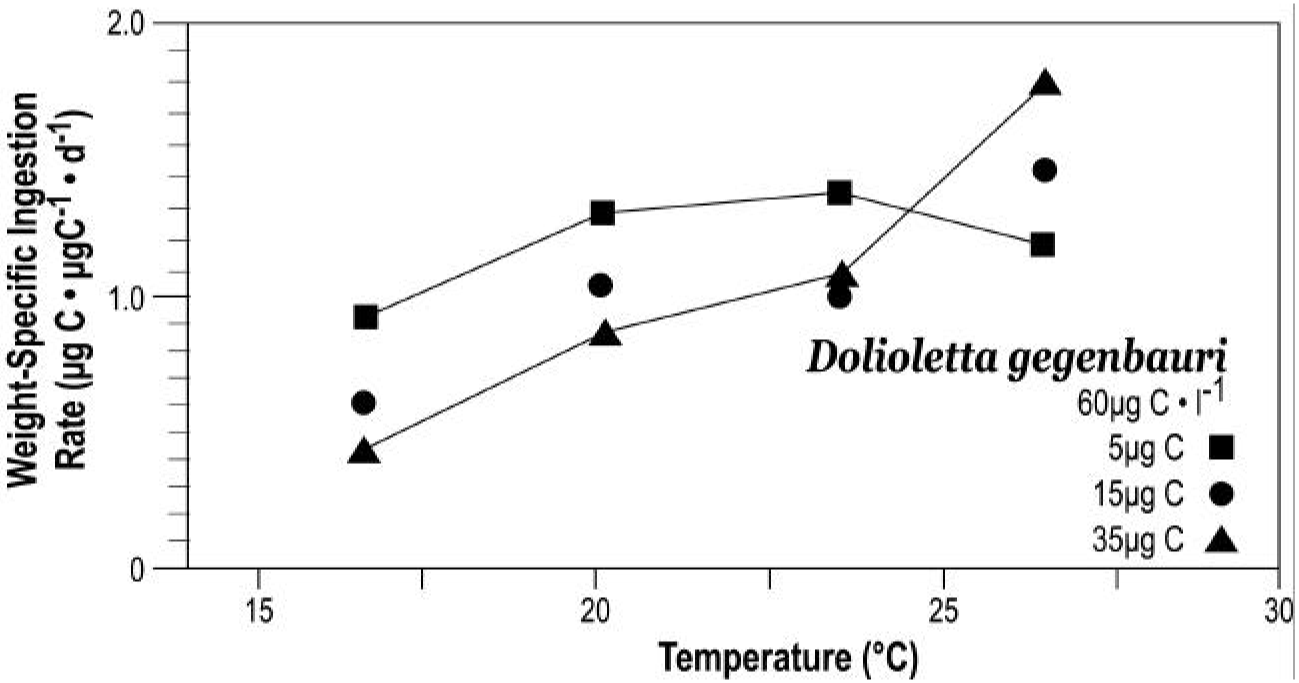

Figure 9. *Dolioletta gegenbauri*. Weight-specific ingestion rates of gonozooids of three different sizes in relation to temperature.

CHAPTER III

ASEXUAL REPRODUCTION OF THE DOLIOLID *DOLIOLETTA GEGENBAURI* Uljanin (Tunicata, Thaliacea)[1]

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[1] Gibson, D.M., G.-A. Paffenhöfer. Submitted to Journal of Plankton Research, 8/21/2000

ABSTRACT

The goal of this research is to enhance our knowledge of the contributions of doliolids to the planktonic community as consumers and secondary producers. The objectives are to quantify asexual reproduction and carbon release rates of *Dolioletta gegenbauri* phorozooids at four food concentrations and four temperatures in order to determine their impact as producers throughout the water column. Although doliolids are abundant in numerous regions of the coastal ocean, and are considered to be major planktonic grazers, data on rates of asexual reproduction are scarce. Laboratory experiments were conducted at 20 ^{o}C at food concentrations of 7, 20, 60, and 160 µg C/l of *Thalassiosira weissflogii* and *Rhodomonas* sp., and at 60 µg C/l at 16.5, 20, 23.5, 26.5 ^{o}C, to quantify the phorozooid release rates of gonozooids, and the amount of carbon released. Results from these experiments suggest that release rates increased at 20 ^{o}C as concentration increases from 7 to 160 µg C/l. Release rates remained similar as the temperature increased from 16.5 to 26.5 ^{o}C at a phytoplankton concentration of 60 µg C/l. Food concentration and temperature has an affect on phorozooid reproductive longevity, size of gonozooids released, and the amount of carbon released asexually. Doliolid reproduction rates are a function of environmental food concentrations and temperatures, and the results imply that doliolids can be important secondary producers in the neritic environment.

INTRODUCTION

In the aquatic realm, asexual reproduction is rare in most phyla. However, is common among protozoa, coelenterata (e.g. Hydroid polyps budding medusae), cladocera (e.g. parthenogenesis of *Daphnia* spp.), and among tunicates (thaliacea). Salps alternate between sexual and asexual reproduction (metagenesis), and doliolids have two asexual and one sexual stage in their lifecycle (Paffenhofer and Gibson, 1999). The nurse produces phorozooids from its cadophore; the phorozooid then produces gonozooids (Godeaux *et al.*, 1998), which is the only sexual stage in the doliolid life cycle. Asexual reproduction results in progeny that are relatively close in size to adults, as compared to eggs, and thus can reproduce more rapidly than via egg production. Another advantage of asexual reproduction is to avoid small juvenile stages that are more prone to predation. Therefore, asexual reproduction results in fewer progeny per day, as compared to the numerous eggs a calanoid copepod may produce daily. For example, Checkley (1980a) reported a maximum of 50 eggs per day for *Paracalanus parvus*.

High abundances of thaliaceans have been observed occasionally (e.g. Deevey, 1952; Atkinson *et al.*, 1978; Pieper and Holliday, 1984; Deibel, 1985; and Koga, 1986). Recent long-term observations showed the evolution over several weeks of doliolids and salp assemblages on the U.S. southeastern continental shelf (Paffenhöfer and Lee, 1987). These blooms could have been the result of fast zooid growth (e.g. Heron, 1972a), high asexual and sexual reproduction rates, low predation, or a combination of all. The impact of thaliacea on primary production is uncertain, particularly because of the limited knowledge on asexual reproduction rates, reproductive longevity, and potential predation on the various zooids. The scarce information on doliolid reproduction (Deibel 1982a; Paffenhöfer and Gibson, 1999), e.g. the number of phorozooids and gonozooids released per day in response to changing phytoplankton concentrations and temperatures, makes it difficult to characterize the effects that an assemblage of doliolids may have on their environment. However, Deibel (1998) proposed that asexual reproduction rates rather than growth rates led to the rapid colonization of neritic regions by doliolids.

The studies we conducted on the feeding and growth rates of gonozooids have enhanced our knowledge of their ability to function at various phytoplankton concentrations and temperatures (Gibson and Paffenhöfer, 2000). The purpose of the present study was to quantify asexual production of gonozooids by phorozooids over their reproductive life at food concentrations and temperatures resembling their natural environment, and to determine if asexual reproduction can lead to rapid colonization of neritic regions. The results of these studies should contribute to improving a model of doliolid activities in planktonic communities on the southeastern shelf of the U.S. (Haskell *et al*., 1999).

METHODS

Dolioletta gegenbauri oozooids, phorozooids, and gonozooids were collected throughout the year from the mid to outer shelf waters along the Wassaw transect off the coast of Savannah GA, onboard the RV Bluefin, at a ship speed of $\leq$ 0.5 kt. A cone net of 200 μm with a 4 L codend, was deployed to collect undamaged doliolids. Onboard ship the contents of the codend were gently floated into 20 L buckets, and up to 20 small undamaged, healthy zooids were separated into 2-L glass jars containing seawater enriched with *Rhodomonas* sp. (a photosynthetic flagellate), to ensure survival on the trip back to the Skidaway laboratory. The 2-L jars were placed on a plankton wheel rotating at 0.2 rpm in an environmental laboratory at 20 ^{o}C, to keep the phytoplankton and zooids in suspension, and at a light-dark cycle of 12h:12h. The zooids were allowed to acclimate to this environment for several days. Each day at least 10% of the water along with aggregated particles, and fecal pellets were removed, and fresh unfiltered seawater was added.

D. gegenbauri were reared through several generations at 20 ^{o}C, the temperature near which they are frequently found on the southeastern shelf ranging from 14 to 22 ^{o}C (Deibel, 1998).

An average food concentrations of the diatom *Thalassiosira weissflogii*, (~ 700 to 1,200µm^3, 80 µg C/mm^3), and the flagellate *Rhodomonas* sp., (~300 µm^3 volume, 160 µg C/mm^3) at 60 µg C/l, which is also comparable to those commonly encountered in nature ranged from ~0.3 to >2 mm ^{3}/l of particulate matter (Paffenhöfer and Lee, 1987a,b).

Phorozooid reproduction and longevity experiments were started with 4 mm phorozooids, which were allowed to acclimate to the experimental temperature and an average food concentration of 60 µg C/l during the initial production of gonozooids. *Rhodomonas sp*. and *Thalassiosira weissflogii* were offered in identical volumetric abundances at the start of each phorozooid reproductive experiment. The concentrations of 7, 20, 60, and 160 µg C/l were chosen for these reproduction experiments to simulate the range of algal concentrations found seasonally on the southeastern continental shelf. Where 20 and 60 µg C/l are often found, and 160 µg C/l is regularly encountered in mature intrusions of the Gulf Stream. The lowest concentration of 7 µg C/l was chosen to determine the response of phorozooid reproduction at extremely low concentrations that may be encountered. The temperatures of 16.5, 20, 23.5, and 26.5 ^{o}C represent the range where *Dolioletta gegenbauri* have been found repeatedly on the southeastern shelf.

Each experiment was designed so that the mean experimental food concentration of *Rhodomonas* sp., and *Thalassiosira weissflogii* was the geometric mean of the initial and final concentration. This was accomplished by conducting preliminary experiments to determine the amount of food remove, and then adjusting the initial concentration accordingly. The first series of reproductive experiments was conducted at 20 ^{o}C and at a mean food concentrations of 7, 20, 60, and 160 µg C/l, with one phorozooid per jar being monitored until death at 10 to 11 mm. Rather than conducting experiments at 390 µg C/l, which showed inhibition in the feeding experiments, a lower concentration of 7µg C/l was used to determine *D.gegenbauri* reproductive capabilities at extremely low food levels. The second series of experiments was conducted at 60 µg C/l and three temperatures, 16.5, 23.5, and 26.5 °C.

Clearance rates were not calculated, but the average phytoplankton reduction per day was measured to ensure food concentration was maintained in the desired range. The same phorozooids were monitored for their entire reproductive life or until death, and the gonozooids released each day were counted and measured a Sony - COHU video systems for SZH microscopes. C/N analysis was conducted on gonozooids reared at 20, 60, and 160 µg C/l (Gibson and Paffenhöfer, 2000). This analysis was performed to determine if carbon content of gonozooids increased as food concentration increased.

Each point on the graphs represents from four to eight reproduction experiments. The Kruskal-Wallis test and the Multiple comparisons test with an alpha = 0.05 were used to determine the similarities and differences of the gonozooid release rates, reproductive longevity of phorozooids, size of gonozooids released, and carbon released in relation to food concentration, and temperatures.

RESULTS

Carbon Content

Gonozooid length vs. carbon content was determined at three phytoplankton concentrations, 20, 60, and 160 µg C/l. Log-linear equations were developed for each concentration. Analysis of covariance indicated that the slopes of each equation were equal. This was supported by a Newman-Keuls test proved that the slopes were not different. There was no significant differences in gonozooid carbon content, at all sizes, found at 20 and 60 µg C/l, and only slightly higher gonozooid carbon values were recorded at 160 µg C/l. Effects of Food Concentration at 20°C

Reproductive Longevity

Phorozooids reproduce asexually by releasing small gonozooids at lengths of 1 to 3.5 mm from a cluster on the ventral peduncle. The term reproductive longevity refers to the period during which phorozooids release gonozooids. The reproductive life span of phorozooids decreased significantly from an average 36 to 24.6 days as the phytoplankton concentration increased from 7 to 20 µg C/l (Fig. 1). Life span was similar at 20 and 60 µg C/l, but then significantly decreased to 7 days at 160 µg C/l.

Phorozooid Release Rates of Gonozooids

As food concentration increased from 7 to 160 µg C/l, release rates increased significantly from 2 to 11 gonozooids/d (Fig. 2). We observed that the size of the cluster of gonozooids on the phorozooid increased with increasing food concentration. This may have contributed to the increased variability in the number of gonozooids released/day (Fig. 2).

Average Size of Newly Released Gonozooids.

As food concentration increased from 7 to 60 µg C/l, a decrease in the average size of gonozooids released ranged from 3.15 to 2.35 mm, followed by a slight increase in the average size of 2.98 mm at 160 µg C/l (Fig. 3). However, these differences were statistically significant.

Phorozooid's Daily and Total Carbon Release Rates

Release rates of carbon as gonozooid by phorozooids are functions of the number and size of gonozooids released per day intergrated over the life span of the phorozooid. The release rates are a measure of the amount of carbon biomass phorozooids return to the planktonic community. The average size of gonozooids released was converted to carbon using the three equations obtained from the C/N analysis, and multiplied by the number of gonozooids released per day.

The amount of gonozooid biomass released per day by phorozooids remained constant as food concentration increased from 20 to 60 µg C/l, and increased significantly at the highest concentration (Fig. 4). However, phytoplankton concentration did not significantly affect the total amount of carbon released over the phorozooids reproductive life at 20 ^{o} C and the 3 different food concentrations (Fig. 5). Decreased longevity of phorozooids at higher concentrations was one factor that contributed to the total amount of gonozooid biomass being similar over the life span.

Temperature Effects at 60 µg C/l

Reproductive Longevity.

As temperature increased from 16.5 to 23.5 ^{o} C, the reproductive life span of the phorozooids remained constant at, 13.8 to 17.3 days, and then decreased significantly to 8.7 days at 26.5°C (Fig. 6).

Phorozooid Release Rates of Gonozooids

With a constant food concentration of 60 µg C/l, increased temperature had little effect on gonozooids releases rates, which ranged from 5.65 to 8.47 gonozooids/day (Fig. 7). Temperature effects were observed at 16.5°C, but they were not statistically significant.

Average Size of Newly Released Gonozooids.

The size of the gonozooids released was affected by temperature (Fig. 8). Gonozooid size was similar at 16.5° to 20°C, increased significantly to 3.02 mm at 23.5°C, and decreased significantly to 1.12 mm at 26.5°C.

Phorozooid's Daily and Total Carbon Release Rates

The amount of gonozooid carbon released per phorozooid increased with increasing temperature from 15.1µg at 16.5° to 47.2 µg C at 23.5°C (Fig. 9), then decreased sharply to 5.26 µg at 26.5°C. All 4 values were significantly different from each other.

The total amount of carbon released over the reproductive life of the phorozooid follows the same trend as the daily carbon release rate (Fig. 10). In this case, however, the total carbon released was similar at 20 and 23.5 ^{o} C, but significantly lower at the lowest and highest temperatures.

DISCUSSION

There is an abundance of information on the somatic growth rates of thaliacea (see Madin and Deibel, 1998 for references), but data on the asexual reproduction rates are scarce (Deibel, 1982a; Paffenhöfer and Gibson, 1999). The occasional occurrence of extraordinary abundances of salps could be attributed to their very high somatic growth rates (Heron, 1972a; Tsuda and Nemoto, 1992), and their estimated population increased due to asexual reproduction (Heron, 1972b). However, unlike salps, doliolid somatic growth rates seldom approached or exceeded a daily doubling in weight (e.g.Gibson and Paffenhöfer, 2000). Our field observations usually revealed populations of the salp *Thalia democratica* near 200 zooids/m^3, yet doliolid populations were near or above 2,000 zooids/m^3 (see Paffenhöfer and Gibson, 1999 for references in their Table1). Which variables contributed to these high doliolid abundances as compared to those of the fast-growing *T.democratica*? In the present study we evaluate whether release rates of gonozooids by phorozooids could make a major contribution to doliolid population growth rate in the presence of limited predation.

Carbon Analysis

In our previous study on feeding and growth of gonozooids, we used one weight to length equation to determine carbon content of gonozooids (Gibson and Paffenhöfer, 2000). This equation was developed from data obtained from gonozooids reared at an average phytoplankton concentration of 60 µg C/l with equal volumetric amounts of *Rhodomonas* sp. and *T. weissflogii*. The only published study adressing the effect of food concentration and temperature on the carbon content of planktonic marine organisms was that of Durbin and Durbin (1992). In this study, the copepod, *Acartia hudsonica's* body weight (carbon) remained stable at food concentrations from 350 to 590 µg C/l, but declined significantly at lower concentrations of 8 to 65 µg C/l. Maximum weight loss was observed to be temperature dependent. We found that as the food concentration increased from 20 to 160 µg C/l, the carbon content of the gonozooids did not change significantly. Without conducting assimilation experiments, we assume that the gonozooids have higher assimilation efficiencies at lower food abundances, i.e. as food concentrations increase assimilation efficiency decreases, allowing them to compensate and maintain relatively constant gonozooid sizes despite changes in food concentration.

Carbon content of gonozooids reared at 7 µg C/l, was not determined assuming that the carbon content would be similar to all others. As observed with A. *hudsonica*, the carbon content would probably have been significantly lower at this food concentration.

Reproductive Longevity

Results from experiments conducted at 20 ^{0}C with 1.0 mm^3/l of *Gymnodinium* and 1.3mm^3/l of *Rhodomonas* showed that the reproductive life span of *Oncaea mediterranea* (Copepoda, Poecilostomatoidea), ranged from 29.1 to 41.7 days, while *Oithona plumifera* (Copepoda, Cyclopoida), averaged 68 and 75 days (Paffenhöfer, 1993). These values were higher than those of *D. gegenbauri*. However, the average reproductive life span of the calanoid *Paracalanus parvus* at a diatom concentration near 25 µg C/l was 10.7 days, and 25 days for *Paracalanus aculeatus*, which are the same range as the phorozooid's. Carlotti *et al*. (1997) showed that at a constant food concentration of 10,000 cells/ml of the haptophycean, *Hymenomonas elongata*, the reproductive lifespan of *Centropages typicus* was 22.3 days at 15°C, and 12.8 days at 20 ^{0}C. Uye (1981) found the same trend with *Acartia clausi*.

In our previous study, (Paffenhöfer and Gibson 1999), phorozooids were offered an average food concentration ranging from 39 to 100 µg C/l, while reproductive lifespan ranged from 8 to 18 days. The food composition varied, the volumetric percentage of respective foods was not always 50:50, and 7 out of the 8 experiments were at average food concentrations ranging from 38.6 to 65.5 µg C/l. In the present study, phorozooids were exposed to food concentrations ranging from 7 to 160 µg C/l at 20 ^{0}C, and their reproductive lifespan, which was longer than the previous study, ranged from 36 to 7 days, respectively. Thus, food quality did not affect the length of the reproductive lifespan.

As the temperature increased from 16.5 to 23.5 ^{0}C with food concentration held constant at 60 µg C/l, the reproductive longevity remained constant, ranging from 13.8 to 17.3 days. We thought that an average food concentration of 60 µg C/l was more ecologically relevant than satiating concentrations, which were used repeatedly in previous copepod studies. Temperatures of 26.5°C reduced the reproductive longevity to 8.7 days. The general trend for doliolids and copepods is that as temperature increases, longevity decreases. In nature doliolids are found from about 16° to 27°C on the southeastern U.S. shelf (Paffenhöfer *et al*., 1991), and occurred abundantly at temperatures below 24°C (e.g. Paffenhöfer *et al*., 1987).

Phorozooid Release Rates of Gonozooids

Only two previous studies have investigated the asexual reproduction of doliolids at 20 ^{0}C. Paffenhöfer and Gibson (1999) found that the average number of gonozooids released per day at an average food concentration of 56.5 µg C/l (n=8) was 11.0/day over 11.2 days. The total number of zooids released per phorozooid lifetime ranged from 83 to 163, being on average 121 + 9.6 (SE), which was similar to the maximum of 129 observed by Deibel (1982a) for a phorozooid surviving for 28 days. These doliolid rates and those presented in the results will be compared to those on egg production rates of several copepod species.

White and Roman (1992) found that egg production of *Acartia tonsa* in Chesapeake Bay is correlated with several factors that vary in relative importance with seasons. Egg production generally increased with temperature up to 27 ^{0}C, then decreased at higher temperatures. It was determined that temperature and microzooplankton abundance, rather than food availability (phytoplankton, chl a), was the best indicator for production. Ambler (1986) determined that, seasonally temperature was the dominant influence on egg production of *A. tonsa*, but that on shorter time scales food quality, quantity, or salinity influenced egg production rates as much as temperature.

Checkley (1980b) found that *Paracalanus parvus* egg production rates ranged from 200-348 eggs/life for phytoplankton concentration range of 12.5 to 63 µg C/l at 18^0C. The daily reproduction as % of body carbon ranged from 17.6 to 46.2, while the total egg production ranged from 176 to 277 (Table 1). These rates were similar to Paffenhöfer (1993), who showed that the total number of eggs produced over the lifetime at 20 ^{0}C for *O. mediterranea* was 274, 254 for *O. plumifera*, and ~250 for *P. parvus*. The daily egg production rate for *O. plumifera* was considerably less than that of *P. parvus*, but due to a reproductive longevity of 71 days, the total egg production was in the same range at 213 (Table 1). For the larger copepod *C. typicus*, Carlotti *et al*. (1997) found that as temperature increased from 15 ^{0}C, egg production over the life of the female was 347 eggs as compared to 235 at 20^0C, and the daily and total production were less than that for *P. parvus*. Uye and Sano (1995) found that daily egg production of *Oithona davisae* (Table 1), increased under eutrophic conditions and with temperatures from 10 to 22 ^{o}C, and then remained constant up to 28 ^{o}C

To make predictions on the contribution of phorozooids to carbon cycling in the neritic environment, not only is the number of gonozooids released important, but also their size and carbon content. In the present study we found that food concentration, and not so much temperature, was the major determining factor in the phorozooid release rates of gonozooids. The average number of gonozooids released per day increased significantly as food concentration increased from 2.4/day at 7 µg C/l, to 11.3/day at 160 µg C/l (Fig.2). As food concentration increased, there was no significant change in the average size of the gonozooids released. The rate of gonozooid production as a % of phorozooid body carbon remained similar, 43.9 and 49%/d, at concentrations from 20 to 60 µg C/l, then increased to 150%/d at the highest concentration. Checkley (1980a) obtained a similar response to food concentration with *P. parvus* (Table 1). Changes in the number of gonozooids released as food concentration increased contributed to the increase in daily reproductive C released.

Release rates remained constant, 5.6 to 8.47 gonozooids/d, when temperature increased from 16.5 to 26.5 ^{0}C. Reproductive rates as % of phorozooid body carbon increased from 26 to 83%/d as temperature increased to 23.5^{0}C. As temperature increased from 16.5 to 23.5^{0}C gonozooid size increased; however, at the highest temperature, very small and non-viable gonozooids were released. The highest temperature seemed detrimental to phorozooid reproduction since reproductive longevity and size of gonozooids released were significantly lower than at all other temperatures. The reproductive rate was only 9.3% of phorozooid body weight per day.

The total number of gonozooids released over the life of the phorozooid for all concentrations and temperature combinations ranged from 46 to 235, the latter from a phorozooid that reproduced for 47 days 20 ^{0}C with a food concentration of 20 µg C/l. The mean number of gonozooids released at 16.5^{0}C and 20^{0}C was 89.4, and 139.2 respectively. The production per lifetime as a % of a phorozooid's body carbon increased to 1270, when concentrations increased from 7 to 20µg C/l, this was assuming that gonozooid carbon content was lower at 7 µg C/l, and remained similar 880 and 960% at the two highest food concentrations at (Table 1). There was a positive correlation between temperature and % body carbon released as gonozooids as temperatures increased from 16.5 to 23.5^{0}C. Lifetime production ranged from 407 to 1180%. However, at the highest temperature, the gonozooids were very small and therefore the lifetime production was the lowest at 80.6% of phorozooid body weight.

The average number of gonozooids released over the life of the phorozooid (longevity · gonozooids released daily), was higher at 20 and 60 μg C/l of food than at 7 and 160 μg C/l; and higher at 20° and 23.5° than at 16.5° and 26.5°C. These results indicate that doliolids can readily reproduce asexually during most months of the year on the middle and outer shelf off the southeastern U.S. where they frequently encounter this range of temperatures and phytoplankton abundances (e.g.Deibel, 1985; Yoder, *et al*., 1979 and 1985; Paffenhöfer *et al*., 1995). Daily reproduction rates of doliolids surpassed those of copepods at similar or higher food concentrations; as did reproduction rates over the entire lifetime.

Energy Transformation

Ingestion rates of individual phorozooids could not be quantified due to the almost constant presence of feeding gonozooids attached to the peduncle or being released during the experimental period. The rate calculated would have therefore been for the entire colony. However, at 60 μg C/l and 16.5 and 26.5 ^{0}C, most of the 6.5 mm phorozooids temporarily lost their large gonozooid cluster for several days at some point during their reproductive life, and had very small non-feeding gonozooids on the peduncle. We compared ingestion rates during this period to the rates of gonozooids at the same temperature, and found that at 16.5 ^{0}C phorozooid ingestion rates were

2.5 times higher, but were 1.7 times lower than gonozooid ingestion rates at 26.5 ^{0}C. Although the ingestion of C/phorozooid/d remained constant between the 10º change in temperature, the carbon released as gonozooids and production as a % of phorozooid ingestion significantly decreased at the highest temperature. However, gonozooid ingestion and growth rates increased between the two temperatures (Table 2). The daily gonozooid release rate, as a% of colony ingestion was 46.6% at 16.5^{0}C, and 13.5% at 26.5^{0}C. The gross growth efficiency for reproduction at 26.5^{0}C was less than a third of that at 16.5^{0}C. At this high temperature, it is probable that the phorozooid's increased metabolic expenditures limited reproductive output (White and Roman, 1992).

Significance of Low Food Concentrations

There are periods when zooplankton production is limited by insufficient food. How a species responds to food limitation will affect its capacity to exploit variations in resource abundances, patterns of occurrence, and contribution so to community structure (Taylor, 1985).

Cladocerans respond to food limitation by curtailing growth and reproduction; in extreme conditions they may produce resting eggs and disappear from the plankton (Taylor, 1985). *P. parvus's* rate of egg production is at times limited by the availability of phytoplankton (Checkley, 1980a), a trend similar to the results Frost (1985) found with *Calanus pacificus*. Studies of copepod reproduction have so far encompassed higher food concentrations. The lowest food concentrations, approximately 12 µg C/l, were used by Checkley (1980a). The ability of copepods to deal with occasional ultra-low food level has yet to be investigated.

In another study of the effect of food limitation on plankton, closely related oligotrich ciliates appeared to respond differently from doliolids at low food concentrations. The majority seemed to require food concentrations near and above 50µg C/l to grow significantly, i.e. to reproduce asexually by division (Montagnes, 1996). The ciliate *Strombidinopsis cheshiri* had a zero growth rate at the low concentrations of 6 µg C/l (Montagnes *et al*., 1996), but was able to grow significantly at food levels above this concentration.

D. gegenbauri phorozooids responded to the lowest food concentration at 7 µg C/l with reduced but long-lasting reproduction (Fig. 1). The size to which the phorozooid grew was not compromised, reproductive longevity was significantly greater than at higher food concentrations, and slightly larger gonozooids were released. However, the number of gonozooid released/day was significantly lower than at all other food concentrations, but the total number of gonozooids released over the lifespan at this concentration was the same as number released at the highest food concentration. Combining all of these factors, doliolids appear to be able to exist and reproduce in environments of very low food abundance and may respond rapidly by increased asexual reproduction once food concentrations increase.

Salps and Doliolids

There may be two reasons contributing to the high doliolid abundances on the S.E. continental shelf, $\geq$ 2000 zooids/m^3 as compared to those of the faster growing *T. democratica*, ~150/m during a thaliacean bloom period: differential predation and asexual reproduction. Information on predation on both, *D. gegenbauri* and *T. democratica* is lacking. The continuously moving salp may have a higher probability of encountering coelenterates and fish as predators (Konchina, 1991) than the occasional-moving doliolid. Salps asexual reproduction appears to be periodic and is difficult to quantify in nature (Heron, 1972b). Doliolids can be studied experimentally due to their limited movements, while salps require larger experimental volumes and some degree of water motion to prevent them from encountering walls, which may hinder feeding. Only with mesocosm experiments then will it be possible to obtain salp asexual reproduction rates at environmental conditions for a more comprehensive comparison and analysis of the life history strategies of salps and doliolids.

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REFERENCES

Ambler, J.W. (1986) Effect of Food Quantity and Quality on Egg Production of *Acartia tonsa* Dana from East Lagoon, Galveston, Texas. *Estuarine, Coastal and Shelf Science*, 23,183-196.

Atkinson, L.P., Paffenhofer, G.-A., and Dunstan, W. M. (1978) The chemical and biological effect of a Gulf Stream intrusion off St. Augustine, Florida. *Bull. Mar. Sci*., 28, 667-679.

Carlotti, F., Rey, C., Javanshir, A., and Nival, S. (1997) Laboratory studies on egg and faecal pellet production of *Centrpages typicus*: effect of age, effect of temperature, individual variability. *J. Plankton Res*., 19, 1143-1165.

Checkley, D.M. (1980a) The egg production of a marine planktonic copepod in relation to its food supply: Laboratory studies. *Limnol. Oceanogr*., 25, 430-446

Checkley, D.M. (1980b) Food limitation of egg production by a marine, planktonic copepod in the sea off southern California. *Limnol. Oceanogr*., 25, 991-996.

Deevey, G. (1952) Quantity and composition of the zooplankton of Block Island Sound, 1949. *Bull. Bingham Oceanogr. Collect*. 13, 120-164.

Deibel, D. (1982a) Laboratory determined mortality, fecundity and growth rates of *Thalia democratica* Forskal and *Dolioletta genenbauri* Uljanin (Tunicata, Thaliacea). *Plankton Res*., 4, 143 - 153.

Deibel, D. (1985) Blooms of the pelagic tunicate, *Dolioletta gegenbauri*: Are they associated with Gulf Stream frontal eddies? *J. Mar. Res*., 43, 211-236.

Deibel, D. (1998) The abundance, distribution and ecological impact of doliolids. In: Bone, Q. (ed.), The Biology of Pelagic Tunicates. Oxford University Press, Oxford, pp. 171-186.

Durbin, E.G. and A.G. Durbin.1992. Effects of temperature and food abundance on grazing and short-term weight change in the marine copepod *Acartia hudsonica*. *Limnol. Oceanogr.*, 27, 361-378.

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

Fryd, M., Haslund, O. H. and Wohlgemuth, O. (1991) Development, growth and egg production of the two copepod species, *Centropages hamatus* and *Centropages typicus* in the laboratory. *J. Plankton Res.*, 13, 683-689.

Gibson, D.M. and Paffenhöfer G.-A. (2000) Feeding and growth rates of the doliolid, *Dolioletta gegenbauri* Uljanin (Tunicata, Thaliacea*). J. Plankton Res.*, 22, 1485-1500.

Godeaux, J., Bone, Q., and Braconnot, J.-C. (1998) Anatomy of Thaliacea. In: The biology of pelagic tunicates. Bone, Q. (ed.), Oxford University Press, Oxford, pp. 1-24.

Haskell, A.E.G., Hofmann, E.E., Paffenhöfer, G.-A. and Verity, P.G. (1999) Modeling the effects of doliolids on the plankton community structure of the southeastern US continental shelf. *J. Plankton Res.*, 21, 1725-1752.

Heron, A.C. (1972a) Population ecology of a colonizing species: The pelagic tunicate *Thalia democratica*. I. Individual growth rate and generation time. *Oecologia*, 10, 269-293.

Heron, A.C. (1972b) Population ecology of a colonizing species: The pelagic tunicate *Thalia democratica*. II. Population growth rate. *Oecologia*, 10, 296-312.

Koga, F. (1986) The occurrence and production of zooplankton in Suo-nada, Western Seto Ireland Sea. *Bull. Nansei Reg. Fish. Res.Lab.*, 20, 91-113.

Konchina, Y.V. (1991) The trophic status of Peruvian fishes in South Pacific oceanic ecosystems. Benguela Trophic Functioning. International Symposium. 8-13 September 1991. Cape Town, South Africa (Abstract)

Madin, L.P. and Deibel, D. (1998) Feeding and energetics of Taliacea. In Bone, Q. (ed.), *The Biology of Pelagic Tunicates*. Oxford University Press, Oxford, pp. 81-103.

Montagnes, D. (1996) Growth responses of planktonic ciliates in the genera *Strombilidium* and *Strombidium*. *Mar. Ecol. Prog. Ser.*, 130, 241-254.

Montagnes, D., Berger, J.D., and Taylor, F. J. R. (1996) Growth rate of the marine planktonic ciliate *Strombidinopsis cheshiri* Snyder and Ohman as a function of food concentration and interclonal variability. *J. Exp. Mar. Biol. Ecol.* , 206, 121-132.

Paffenhöfer, G.-A. and Lee, T.N. (1987a) Development and persistence of patches of Thaliacea. *S. Afr. J. Mar. Sci.*, 5, 305-318.

Paffenhöfer, G.-A. and Lee, T.N. (1987b) Summertime Gulf Stream intrusions into southeast U. S. shelf waters in 1981: Changes in distribution and abundance of particulate matters. *Prog. Oceanogr*. 19, 373-401.

Paffenhöfer, G.-A., Sherman, B.K. and Lee, T.N. (1987) Summer upwelling on the southeastern continental shelf of the U.S.A. during 1981: Abundance, distribution and patch formation of zooplankton. *Prog. Oceanogr*. 19, 403-436.

Paffenhöfer, G.-A. (1993) On the ecology of marine cyclopoid copepods (Crustacea, Copepoda). *J of Plankton Res***.,** 15, 37-55.

Paffenhöfer, G.-A., Stewart, T.B., Youngbluth , M.J., and Bailey T.G. (1991) High – resolution vertical profiles of pelagic tunicates. *J.Plankton Res.,* 13, 971-981.

Paffenhöfer, G.-A. and Gibson, D.M. (1999) Determination of generation time and Asexual fecundity of doliolids (Tunicata, Thaliacea). *J. Plankton Res.*, 21, 1183-1189.

Pieper, R.E., Holliday, D.V. (1984) Acoustic measurements of zooplankton distributions in the sea. *J. Cons. Int. Explor. Mar.*, 41, 226-238.

Taylor, B.E. (1985) Effects of food limitation on growth and reproduction of *Daphnia. Arch. Hydrobiol. Beih. Ergebn. Limno*., 21, 285-296.

Tsuda, A., and Memoto, T. (1992) Distribution and growth of salps in a Kuroshio warm-core ring during summer 1982. *Deep-Sea Res.*, 39 (Suppl. 1), S219-S229.

Turner, J.T., Tester, P.A., and Hettler, W.F. (1985) Zooplnnkton feeding ecology: A laboratory study of predation on fish eggs and larvae by the copepods *Anomalocera ornata* and *Centropages typicus*. *Mar. Biol.,* 90, 1-8.

Uye, S. (1981) Fecundity studies of the neritic calanoid copepods *Acartia clausi* Giesbrecht and *A. steurei* Smirnov: a simple empirical model of daily egg production. *J. Exp. Mar. Biol. Ecol.*, 50, 255-271.

Uye, S. and Sano, K., (1995) Seasonal reproductive biology of the small cyclopoid copepod *Oithona davisae* in a temperate eutrophic inlet. *Mar. Ecol. Prog. Ser.*, 118, 121-128.

White J.R. and Roman M.R. (1992) Egg production by the calanoid copepod in the mesohaline Chesapeake Bay: the importance of food resources and temperature. *Mar. Ecol. Prog. Ser.,* 86, 239-249.

Yoder. J.A., Atkinson, L.P., Lee, T.N. (1981) Role of Gulf Stream frontal eddies in forming phytoplankton patches on the outer southeastern shelf. *Limnol. Oceanogr.,* 26, 1103-1110

Yoder, J.A., Atkinson, L.P., Bishop, S.S., Blanton, J.O., Lee, T.N., and Pietrafesa, L.J. (1985) Phytoplankton dynamics within gulf Stream intrusions on the southeastern United States continental shelf during summer 1981. *Cont.Shelf Res.* 4, 611-635.

Table I Daily and Total reproduction as % body carbon for copepods and phorozooids

Species	Temperature ^{o}C	Food Conc. mg C/l	Rate /d	Longevity (days)	Daily reprod. % of Body C	Total reproduction as % of Body C	References
			eggs/d				
Paracalanus parvus	18	12.5	20	10	17.6	176	Checkley 1980a
		63	58	5	46.2	277	
Centropages typicus	15	1400	15.59	22.25	4.4	99	Carlotti et al. 1997
	20	"	15.16	12.77	4.32	55.4	
Oithona plumifera	20	varied	4	71	3	213	Paffenhofer 1993
Oithona davisae	10	eutrophic	NA	Not Availible	8	NA	Uye and Sano 1995
	15				20		
	20				35		
	25				38		
	28				38		
			gonozooids/d				
Dolioletta gegenbauri	20	56.5	11	11	64.8	960	Paffenhöfer and Gibson1999
Dolioletta gegenbauri	20	7	2.4	36	NA	NA	Present study
	20	20	5.48	24.6	43.9	1270	
	20	60	8.08	17.3	49	880	
	20	160	11.28	7	150	960	
	16.5	60	5.65	15.4	29	40.7	
	20	60	8.08	17.3	49	880	
	23.5	60	7.97	13.8	83.1	1180	
	26.5	60	8.47	8.7	9.3	80.6	

Table II Clearance and ingestion rate comparisons for *D. gegenbauri* phorozooids, gonozooids, and phorozooid production as % of ingestion.

Concentration µg C/l	Temperature °C	Clearance Rate ml/zooid/d		Ingestion Rate µg C/zooid/d		Carbon Released/d (phorozooid)	Production as % of Ingestion (phorozooid)
		Phorozooid	Gonozooid	Phorozooid	Gonozooid		
60.0	16.5	566	293	33.9	15.6	15.11	46.6
60.0	26.5	649	1400	38.9	62.0	5.26	13.5

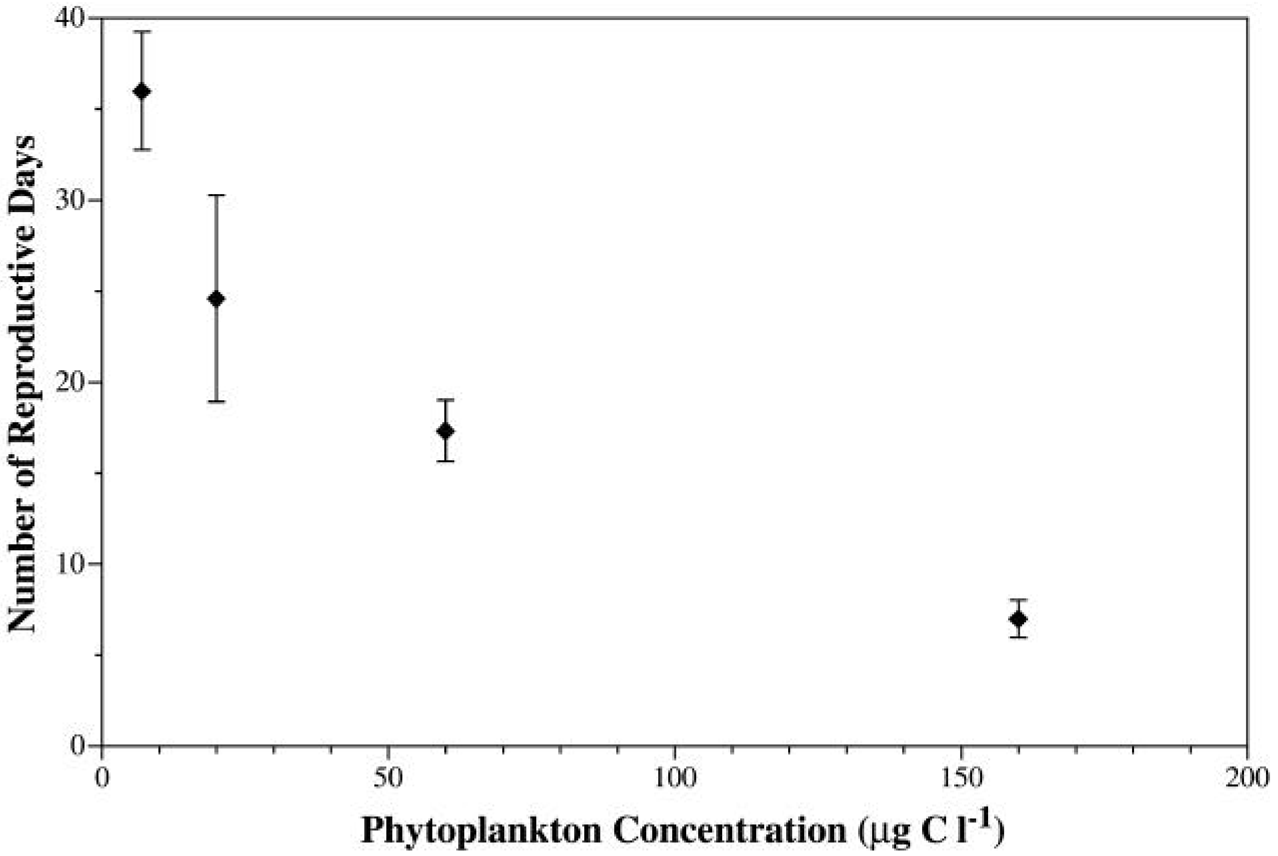

Figure 1 Mean (+ SE) number of phorozooid reproductive days (longevity) versus phytoplankton concentration (µg C l $^{-1}$), at 20 ^{0} C. 20 µg C/l = 60 µg C/l, all others are significantly different, using Kruskall Wallis and Multiple Comparison tests.

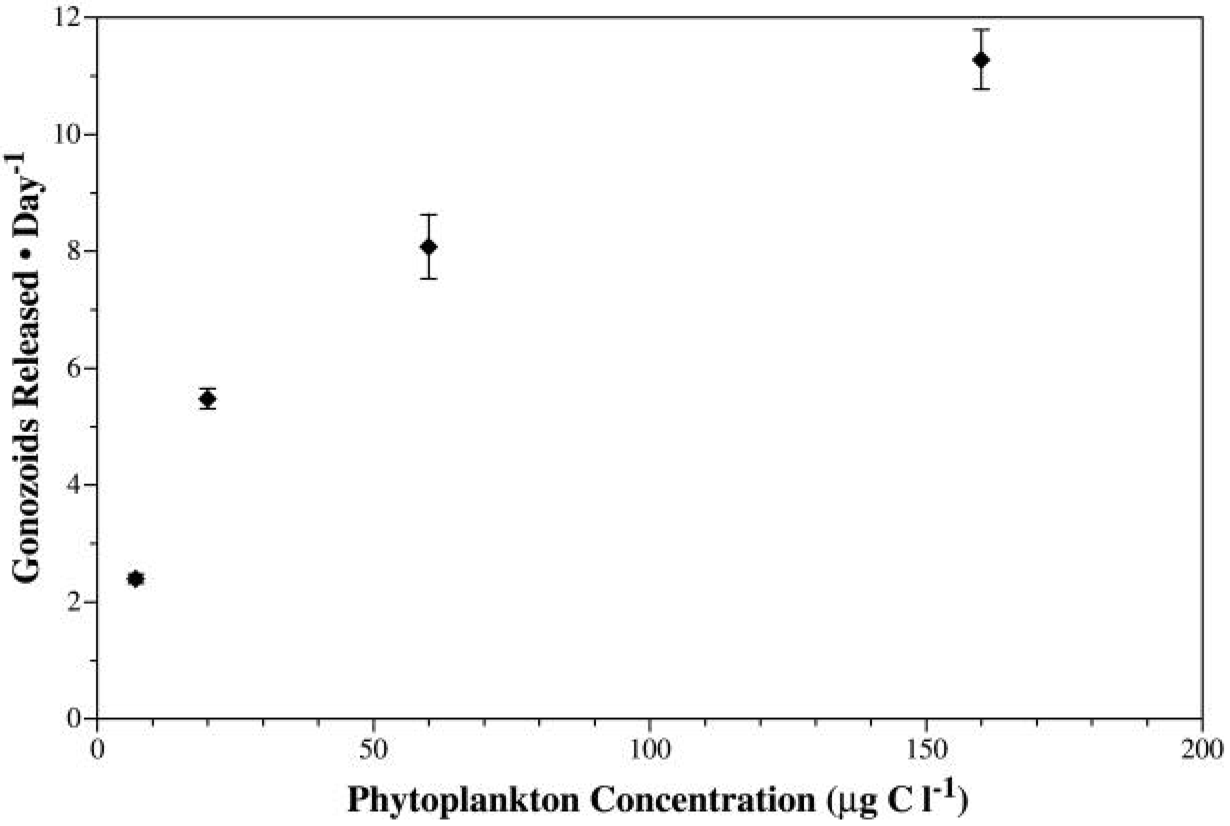

Figure 2 Mean (+ SE) number of gonozooids released per day by a phorozooid versus phytoplankton concentration (µg C l $^{-1}$), at 20 ^{0} C. All rates are significantly different.

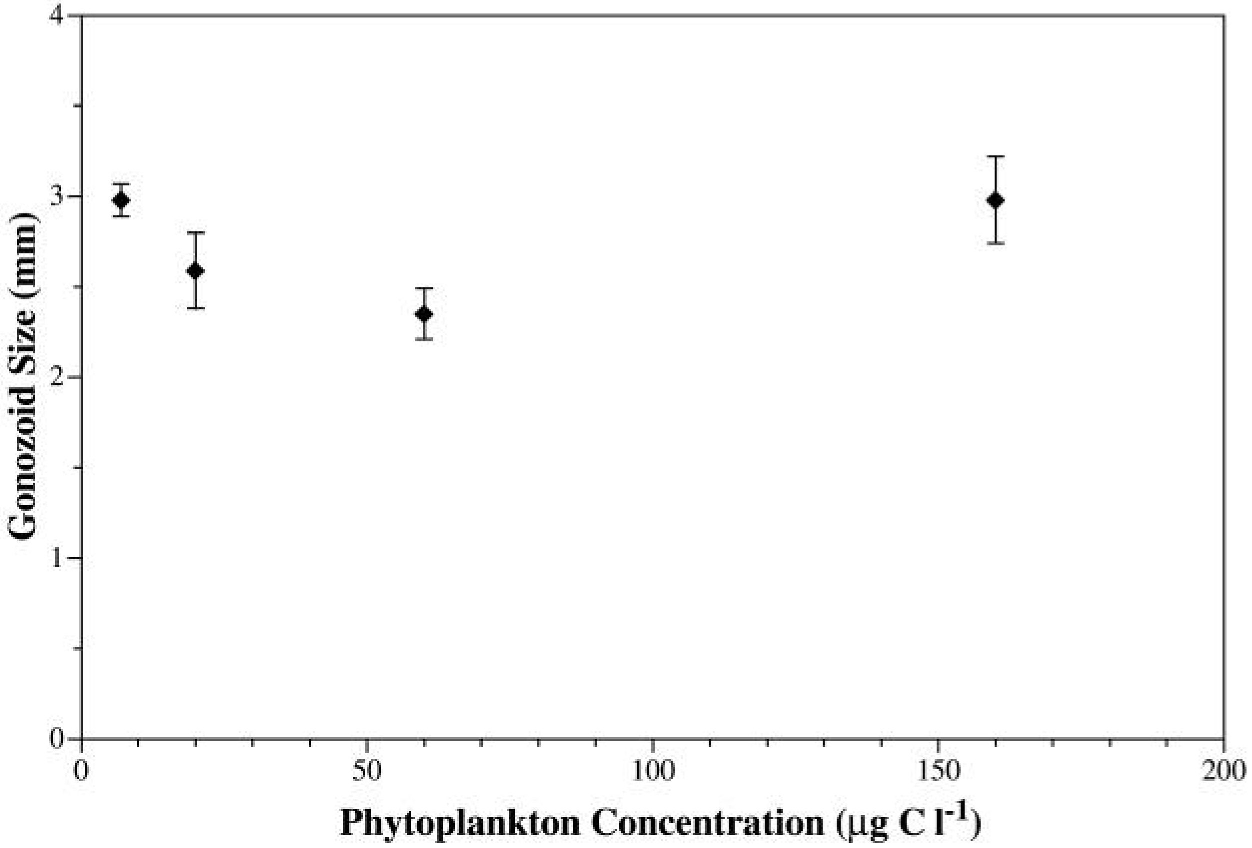

Figure 3 Mean (+ SE) size of gonozooids released per day versus phytoplankton concentration (μg C l $^{-1}$), at 20 ^{0} C. Mean sizes are not significantly different.

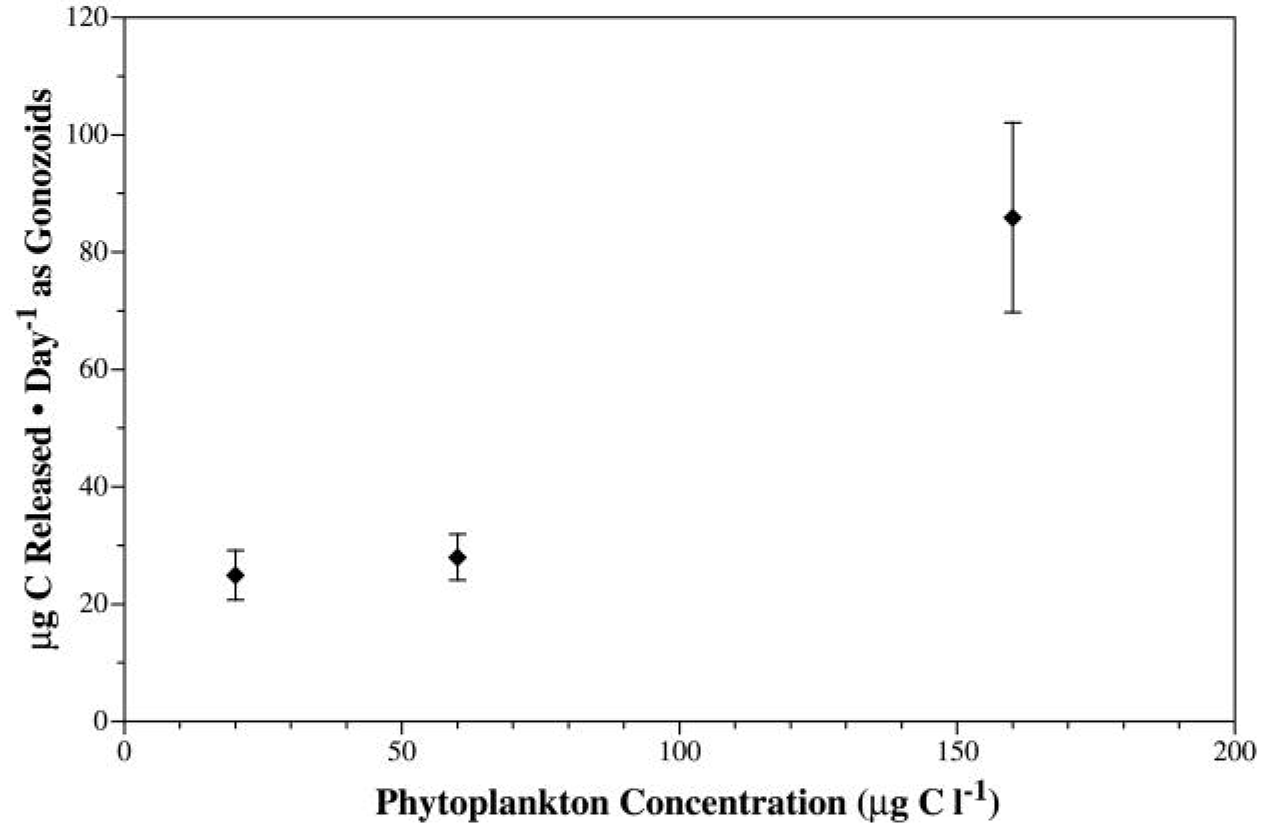

Figure 4 Mean (± SE) amount of carbon released as gonozooids per day versus phytoplankton concentration (µg C l $^{-1}$), at 20 ^{0} C. 7 = 20 = 60 µg C/l, 160 µg C/l is significantly greater than other values.

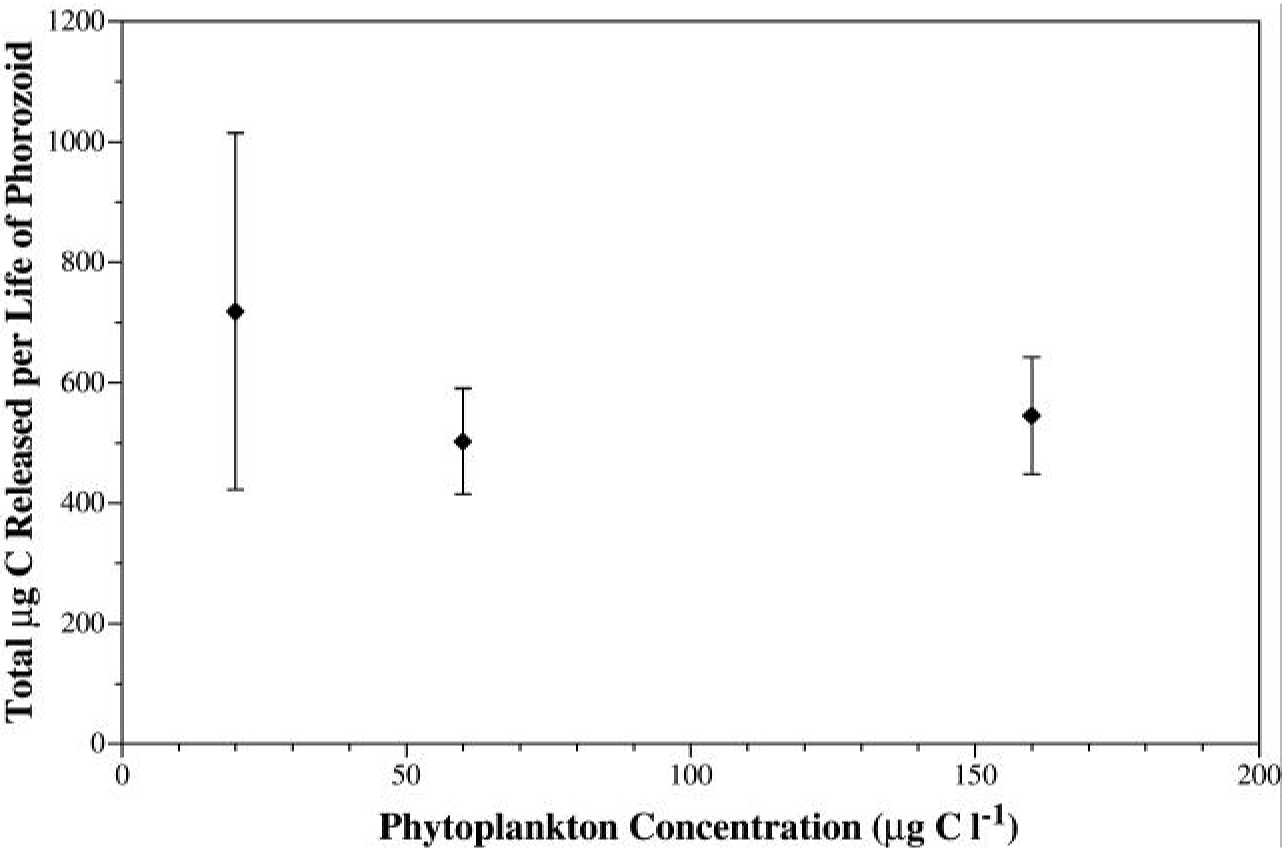

Figure 5 Mean (± SE) amount of carbon released over the life of the phorozooid versus phytoplankton concentration (µg C l $^{-1}$), at 20 ^{0} C. Mean release rates are not significantly different, however, the rate at 20 µg C l $^{-1}$ has a higher SE due to one phorozooid surviving for 47 days and releasing a total of 235 gonozooids.

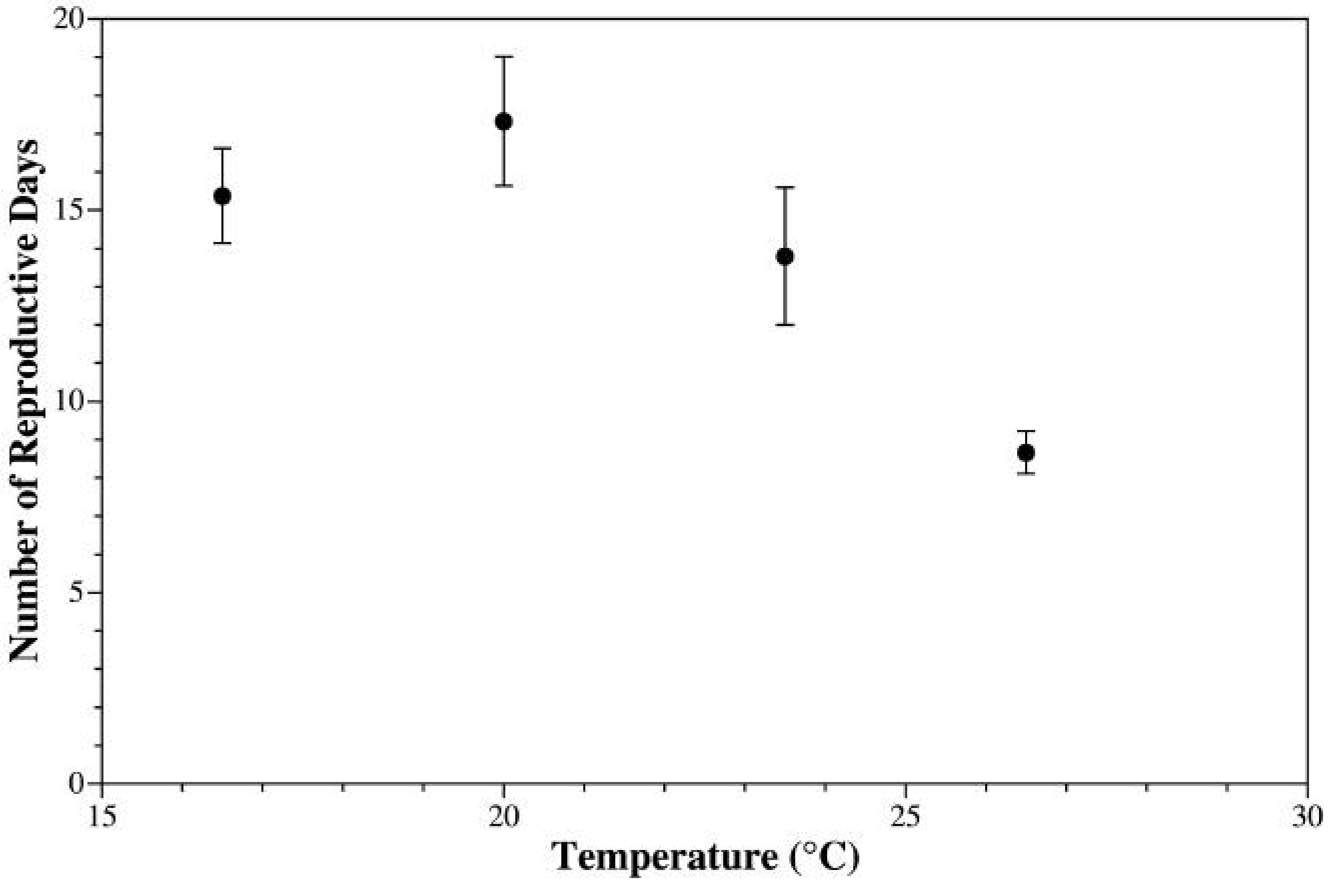

Figure 6 Mean (± SE) number of phorozooid reproductive days (longevity) versus temperature (^{0} C), at a food concentration of 60 µg C l^{-1}. 7 = 20 = 60 µg C/l, 160 µg C/l is significantly lower than other values.

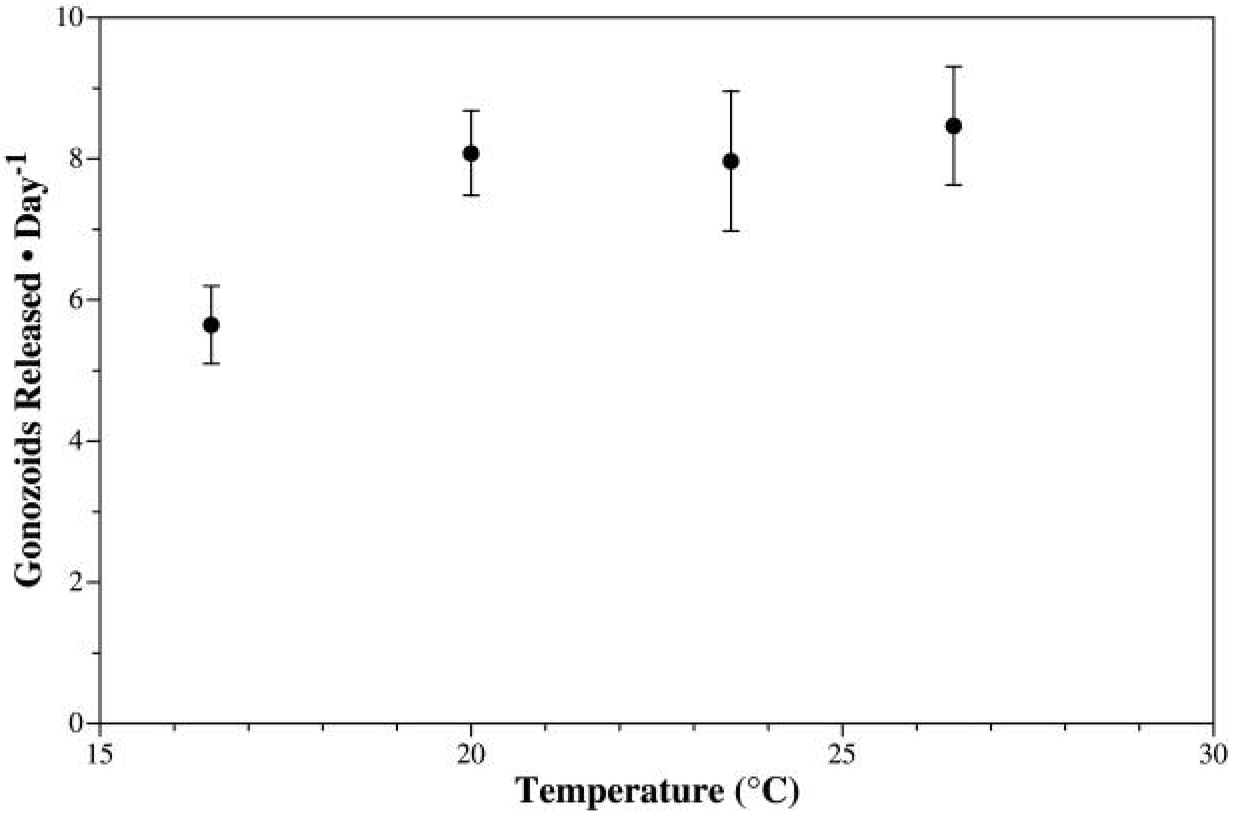

Figure 7 Mean ($\pm$ SE) number of gonozooids released per day by a phorozooid versus temperature (^{0} C), at a food concentration of 60 µg C l^{-1}. Mean release rates are not significantly different.

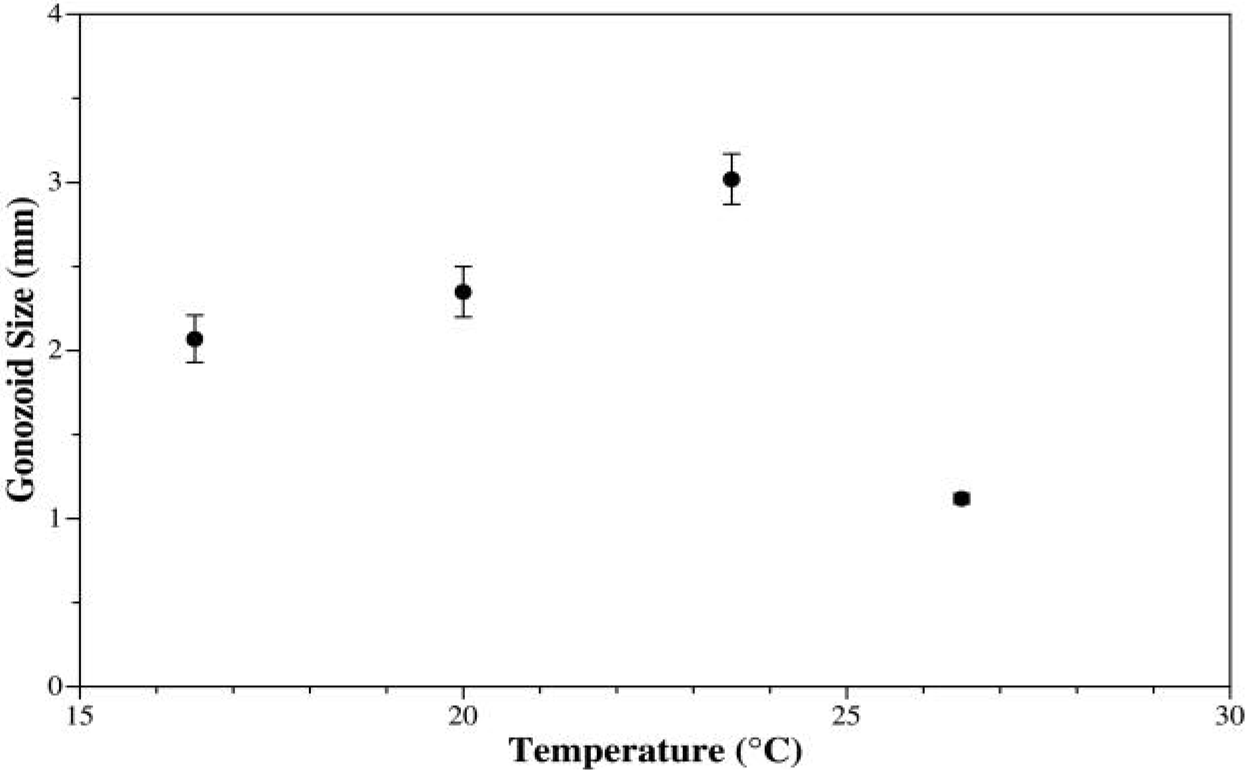

Figure 8 Mean (+ SE) size of gonozooids released per day versus temperature (^{0} C), at a food concentration of 60 µg C l $^{-1}$. 7 = 20,µg C l $^{-1}$, 60 µg C/l is significantly higher than other values, and 160 µg C/l is significantly lower than other values.

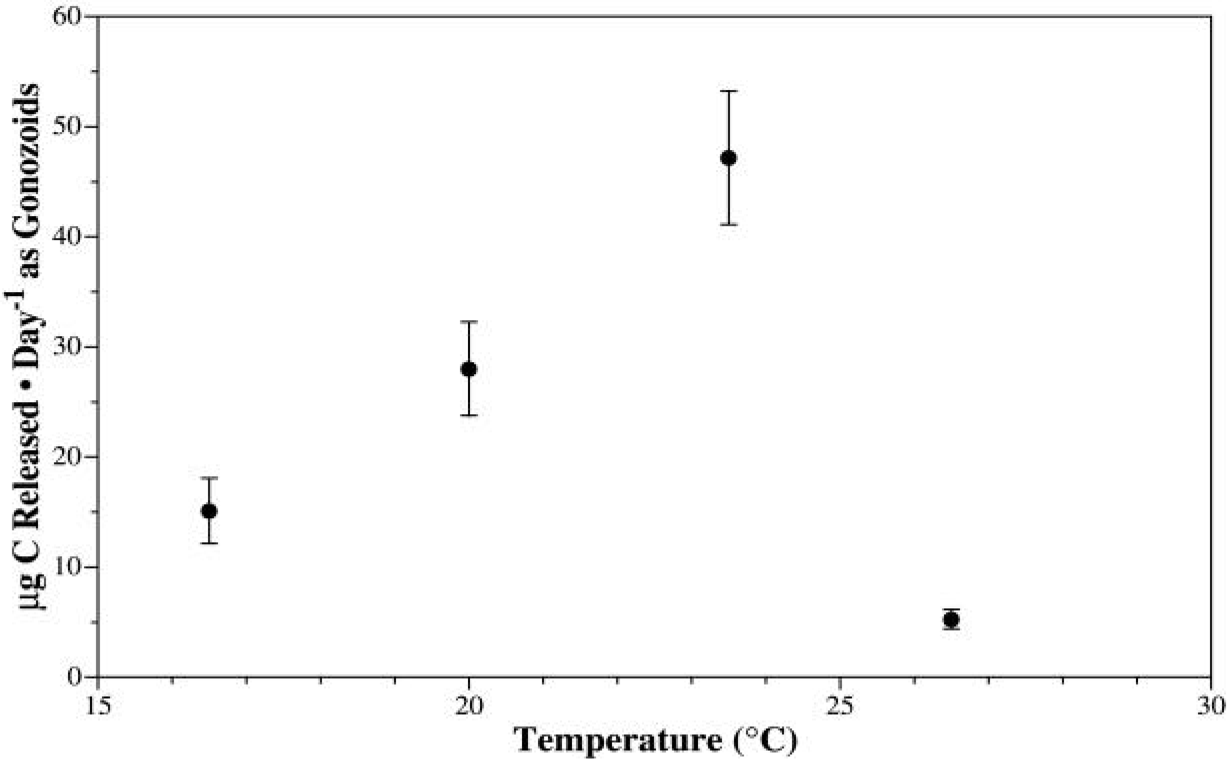

Figure 9 Mean (+ SE) amount of carbon released as gonozooids per day versus versus temperature (^{0} C), at a food concentration of 60 µg C l $^{-1}$. Mean rates are all significantly different.

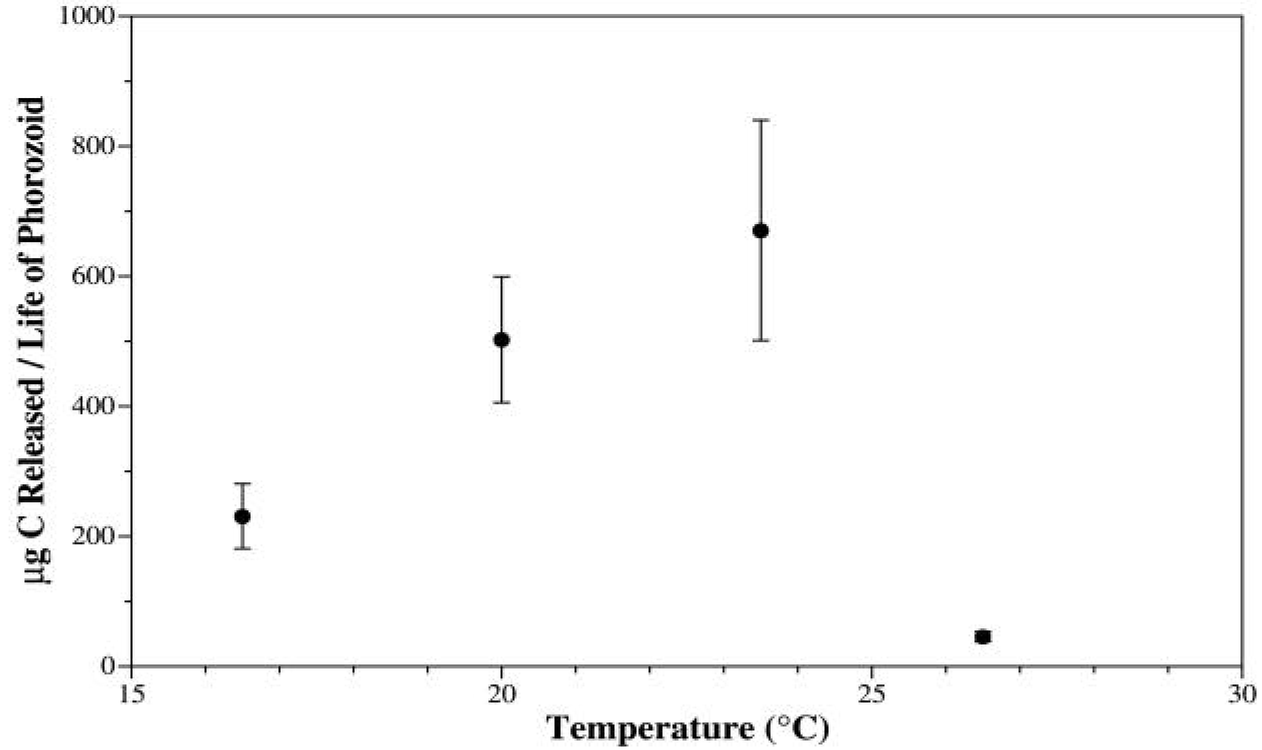

Figure 10 Mean ($\underline{+}$ SE) amount of carbon released over the life of the phorozooid versus temperature (^{0} C), at a food concentration of 60 μg C l $^{-1}$. 7 ≠ 20μg C l $^{-1}$, 20 = 60μg C l $^{-1}$, and 160 μg C/l is significantly lower than other values.

CHAPTER IV

FEEDING RATES OF THE DOLIOLID, *DOLIOLETTA GEGENBAURI* ULJANIN (TUNICATA, THALIACEA), ON THE DIATOM *RHIZOSOLENIA ALATA*[1]

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[1] Gibson, D. M. and G.–A. Paffenhöfer. To be submitted to Journal of Plankton Research.

ABSTRACT

We hypothesized that the presence of large diatoms would affect the feeding of gonozooids of *D. gegenbauri* on smaller, co-occurring cells. To test this hypothesis, we measured the clearance and ingestion rates of 3.5 to 5.5 mm *Dolioletta gegenbauri* gonozooids on phytoplankters of different sizes in a mixture of, *Rhodomonas sp*. (*Rho*.) 8µm; *Thalassiosira weissfloggii* (*Tw*) 12µm; and *Rhizosolenia alata* (*R. alata*) 49 µm cell diameter, at 20 ^{o}C.. The objectives of this study were to first, quantify the rates at which each size zooid would feed at three concentrations (21, 46, 100µg C/l) of *R. alata*, with *Rho* and *Tw* kept at a constant concentration of 60 µg C/l; second, to determine how these rates would affect the clearance rates of the co-occurring *Rho* and *Tw* cells; and third, to compare these rates to the rates from experiments where gonozooids were offered 60 µg C/l of *Rho* + *Tw* only. The clearance rates of *R. alata, Rho*, *Tw*, and the *Rho+Tw* combination were not significantly different at the low concentration of *R. alata* for all gonozooid sizes. However, some differences were found in clearance rates at the medium and high concentrations**.** The presence of *R. alata* inhibited ingestion rates of smaller cells as compared to our previous study; however, these large diatoms increased the percentage of carbon ingested/day. The results from these experiments suggest that as *Dolioletta gegenbauri* gonozooids increased from 3.5 to 5.5 mm, their ability to ingest *R. alata* increased. Small and medium sized gonozooids can ingest large and small cells at similar rates at all concentrations, while at highest concentration of *R. alata* the larger gonozooid's clearance rates on the large diatom, reduced the grazing pressure on smaller cells. The ingestion of large diatoms increases the amount of carbon ingested per gonozooid per day, and may increase growth and reproduction rates.

INTRODUCTION

Several studies utilizing natural particles and particles of different sizes, have been conducted to determine feeding rates and retention efficiencies of salps, doliolids, and copepods (Paffenhöfer and Knowles, 1978; Harbison and McAlister, 1979; Deibel 1982, 1985; Paffenhöfer, 1984; Deibel and Paffenhöfer, 1988; Crocker *et. al*, 1991; Tebeau and Madin, 1994). One study, focused on determining the particle retention spectra of mucous nets of three salp species as a function of grazing rates and particles of different sizes, concluded that smaller salps can retain a greater fraction of small particles (Harbison and McAlister, 1979). Deibel (1982) found that there was no significant difference in mean grazing rates of *Thalia democratica* and *Dolioletta gegenbauri's* gonozooids, phorozooids and oozooids feeding on the flagellate *Isochrysis galbana* and the dinoflagellate *Peridinium trochoideum*. Another study revealed that clearance rates of net captured *Thalia democratica*, offered natural particles, were slightly lower than rates measure in the laboratory, while ingestion rates of net captured salps were slightly higher than rates determined in the laboratory (Deibel, 1985). Deibel and Paffenhöfer (1988) observed that doliolids were able to manipulate and compact larger *Ceratium sp*. before ingestion. Zooids of *Dolioletta gegenbauri* efficiently ingested radiolabelled bacteria < 5um, and diatoms > 100um (Crocker *et. al*, 1991). Tebeau and Madin (1994), determined that trophozooids and phorozooids with lengths from 1.1 to 2.9 mm on the nurse colony are capable of filtering 1.0 - 2.5 um particles at similar rates, but retention efficiencies of the 1.0 um particles were < 100%.

Studies on the rate of doliolids feeding under simulated or real environmental conditions require that food composition, abundances, and size distribution resemble that in the natural environment (Paffenhöfer, 1984). Therefore, the goal of the present study is to determine clearance and ingestion rates of gonozooids from 3.5 to 5.5 mm length, with increasing abundances of *R. alata*, a diatom predominant on the southeastern continental shelf throughout most of the year, and compare these results to previous observations.

METHODS

Dolioletta gegenbauri oozooids, phorozooids, and gonozooids were collected throughout the year from the mid shelf waters along the Wassaw transect off the coast of Savannah GA, onboard the RV Blue Fin at a ship speed of ≤ 0.5 kt. A cone net of 200 μm mesh with a 4 L codend was deployed to collect undamaged doliolids. The contents of the codend were gently floated into 20 L buckets, and up to 20 small, undamaged, healthy zooids were separated into 2-L glass jars containing seawater enriched with *Rhodomonas* sp. was added to ensure survival on the trip back to the Skidaway laboratory. The 2-L jars were then placed on a plankton wheel rotating at 0.2 rpm in a light-dark cycle of 12h: 12h at 20 ^{o}C, to keep the phytoplankton and zooids in suspension. The zooids were allowed to acclimate to this environment for several days. Each day at least 10% of the water, aggregated particles, and fecal pellets were removed, and fresh unfiltered seawater was added.

Feeding experiments were conducted with gonozooids at 20 ^{o}C, and three mean concentrations of 21, 46, and 100 μg C /l of *Rhizosolenia alata* (0.8×10^{6} μm ^{3} cell volume, 13.9 μg C/mm^{3}, Strathmann 1967). A constant concentration of 60 μg C /l of the diatom *Thalassiosira weissflogii,* (~ 700 to 1,200 μm ^{3}, 80 μg C/mm^{3}), and the flagellate *Rhodomonas* sp., (~300 μm^{3} volume, 160 μg C/mm^{3}) was maintained for all experiments. Each experiment was designed so that the mean experimental food concentration of *Rhizosolenia alata*, *Rhodomonas* sp., and

Thalassiosira weissflogii was the geometric mean of the initial and final concentration. This was achieved by utilizing the known clearance rates of gonozooids at a particular size (Gibson and Paffenhöfer, 2000), the appropriate number of zooids was added to each experiment to insured that the initial food concentration would not be reduced by more than 30%. Measurements were made of the body length and branchial aperture (the opening to the branchial cavity), at the beginning and end of each experiment using a Sony video system (Gibson and Paffenhöfer, 2000). Clearance rate experiments were conducted using gonozooids that were initially 3 mm until maturity at 6 mm. Individual, cell clearance rates were measured by using an inverted microscope to determine differences between food concentrations at the beginning and end of the experiment. Phytoplankton growth constants in the controls, gonozooid clearance (ml/zooid/day), and ingestion (µg/C/zooid/day) were calculated using the equations of Frost (1972), and Harbison and Gilmer (1976). Each point on the graph represents from three to thirteen feeding experiments Fig. 1. The Kruskal-Wallis test and the Multiple Comparisons test with an $\alpha = 0.05$, were used to determine the differences of clearance rates between food type, zooid sizes, and food concentrations.

RESULTS

Branchial Aperture Measurements

In order to determine the ability of 3.5 to 5.5 mm gonozooids to ingest *R. alata* (average 49 µm cell width x 450 µm cell length), measurements were made of the zooid's length and branchial aperture. The aperture diameter ranged from 0.91 to 1.65 mm, with an average length to aperture ratio of 3.91. The largest *R. alata* cell measured was 50 X 500 µm, which may be small enough to be ingested by the smallest gonozooid.

Clearance Rates

Clearance rates of gonozooids ranging from 3.5 to 5.5 mm feeding on *R. alata* at 21 µg C/l, and *Rho sp*. and *T. w* at 60 µg C/l, increased with increasing gonozooid size. The rates ranged from 300 to 870 ml/d for co-occurring *R. alata*, *Rho sp*. and *T. w*, and were not significantly different at each doliolid size. However, these rates were significantly different, as the gonozooid size increased, and were slightly higher than the rates determined from our previous experiments (Fig. 1A).

When the concentration of *R. alata* was increased to 46 µg C/l, *R. alata* clearance rates were significantly lower than the rates of the smaller cells for the small gonozooids, but similar rates were found for all cells for the medium sized gonozooids. However, the clearance rates for the 5.5 mm gonozooid on *R. a*lata were not significantly different from the *Rho + T.w* combination from previous experiments, but it appeared that the high clearance rate on *R. alata* may have resulted in reduced rates on the co-occurring smaller cells (Fig. 1B).

An increase of *R. alata* cells to 100 µg C/l, with co-occurring *Rho and T.w* concentrations remaining the same, resulted in an increase in clearance rates with increasing zooid length on all cells except *T.w*. Clearance rates on all co-occurring cells were not significantly different for the 3.5 mm gonozooids. However, these rates were lower than rates on *Rho + T.w* from our previous study. The medium and large zooid's clearance rates on *T.w* were significantly lower than all other cells, but the rates for co-occurring *R. alata* and *Rho* were not significantly lower than rates on *Rho + T.w* (Fig. 1C).

An overview of clearance rates for all zooid sizes, and particle combinations and concentrations, reveals a decrease in clearance rates with increasing *R. alata* abundance for the medium and large gonozooids. Large zooids removed *R. alata* at a significantly higher rate than the co-occurring *Rho and T.w* in the 46 and 100 µg C/l treatments. In contrast, small gonozooid clearance rates on all cells are similar at these concentrations of *R. alata.*

Ingestion Rates

There was a positive correlation between gonozooid size and ingestion rates (µg C/zooid/d) *of R. alata, Rhodomonas sp*. and *T. weissflogii* at each *R. alata* concentration. At 21µg C/l *of R. alata*, the amount of carbon ingested by the small and medium size zooids is similar for *R. alata* and *Rhodomonas sp.*, but is significantly higher than the rate for *T.w*. The largest gonozooid has significantly different rates of ingestion for all particles (Fig. 2A).

The ingestion rates on *R. alata* cells significantly increase with increasing zooid size at 46 and 100µg C/l (Fig. 2B and C), and slight increases in ingestion for *Rho* and *T.w* as zooids increase from 3.5 to 4.5 mm. Except at 46 µg C/l, the high ingestion rate of the large gonozooid significantly reduces the ingestion rate of *Rho and T.w*. (Fig. 2B). Ingestion rates of *R. alata* at 100µg C/l are significantly higher than co-occurring food particles for all zooid sizes (Fig. 2C).

Weight-specific ingestion for the 3.5 mm (~ 8 µg C) gonozooids were160-170 %, as the concentration of *R. alata* increased. Significant differences in ingestion among food concentrations were observed for the 15µg C zooid, (155 to 256%), and for the 24µg C gonozooids, (140 to 190%) (Fig. 3). Maximum rates were found at 100µg C/l with no obvious relation to body weight.

However, the minimum rates were at the lowest concentration, decreasing with increasing body weight.

DISCUSSION

Clearance Rates

The present study focuses on the clearance rates of *D. gegenbauri* on *R. alata, Rhodomonas sp*. and *T. weissflogii,* representing the particle size distribution encountered in upwelled waters on the southeastern shelf of the U.S.A. (Paffenhofer and Lee, 1987). Clearance rates of *Rho* and *T. w*. from present experiments were compared to *Rho+Tw* from previous experiments (Gibson and Paffenhöfer, 2000), to determine if the ingestion of large cells would alter the clearance rates of smaller cells, and if the previous bi-culture experiments would accurately represent feeding rates on cultures of various particle sizes.

Table I summarizes clearance rates of doliolids and salps feeding on natural and large particles. Feeding studies of *D. gegenbauri* gonozooids, phorozooids, and oozooids from 2 to 12 mm were conducted by Crocker *et. al* (1991) on bacteria (0.2 – 5µm), and *Nitzschia seriata* (7.8 x 102um), at 12-14 ^{o}C in 275 to 500 ml jars for 1-5 hours. Gonozooid mean clearance rates, 105 ml/d, increased with size, and were similar for both bacteria and diatoms, which indicates equal collection efficiency. These rates were considerably lower than rates from our present and previous studies. The present study was conducted at 20 ^{o}C in 2 L jars for up to 24 hours. Differences in these rates could be explained by an increase in temperature, which was found to have a major influence on clearance rates (Gibson and Paffenhöfer, 2000); larger vessel volumes may decrease the chances of zooids encountering walls to disrupt feeding; and longer experimental times may allow for diurnal variability (Deibel 1982).

Deibel (1985) studied the clearance and ingestion rates of *Thalia democratica* on naturally occurring particles, and determined that *T. democratica* can consume particles from 20 to 30 µm ESD (Equivalent Spherical Diameter) when there is a concentration peak from 10 to 20 µm ESD. The upper size limit of particles ingested by *Thalia* is not known due to constraints by the mouth diameter, which Deibel measured to be 45 – 65 µm diameter. Deibel (1982b) found in a laboratory study that, *T. democratica and D. gegenbauri* were capable of ingesting the dinoflagellate *Peridinium trochoideum* (16-18 µm diameter), but not the *Rhizosolenia sp.* (5-60 µm diameter, 180-350µm length). However in the present study, it was determined that at low concentrations of *R. alata*, the clearance rates of all cells were not significantly different from the rates of the previous study at similar concentrations. As the concentration of these large cells increased, the clearance rates of the small cells were lower than the rates from the previous study, especially for the larger gonozooids. These results suggest that in mixed sized phytoplankton cultures, gonozooids can ingest different sized cells at similar rates, and that large gonzooids have the ability to clear the larger, numerically abundant cells, while the clearance rates of small cells are significantly reduced.

Effect of Branchial Aperture Size on Clearance Rates

Personal observations by Paffenhöfer (unpublished) determined that doliolids ceased feeding when large particles touched the branchial lobes. The branchial aperture was measured to determine if large *R. alata* cells would disrupt the feeding of the smallest gonozooids. The aperture diameter increased from 0.91-1.45 mm as gonozooids increased from 3.5-5.5 mm length. The average *R. alata* cell dimensions were 49µm width x 450µm length. The aperture of the smallest gonozooid appeared to be large enough to ingest these cells.

The clearance rates of these large cells increased at each concentration as the zooid length and aperture size increased. Personal observation of a few large gonozooids ingesting *R. alata*, revealed that the intake at the aperture did not disrupt feeding, but handling of these large cells at the mouth took some effort. To ingest the cell, the gonozooids would repeatedly rotate their entire body to position the cell into the mouth. Deibel and Paffenhofer (1988) filmed *D. nationalis* ingesting large *Ceratium spp*. cells in 6 cyclic reversals of the mucous cord from the ciliated funnel to the mouth. The gill cilia ceased beating when large or noxious particles touched the mouth, but some large particles were wrapped in a mucous cocoon and ingested. This may explain why the smaller gonozooids had lower clearance rates, why all rates were lower at the highest concentration, and why the ingestion of larger cells reduced the clearance of co-occurring smaller particles.

Ingestion Rates

Ingestion rates of thaliaceans generally increase with increasing food concentration, (Deibel, 1982 for *T. democratica* and *D. gegenbauri*, and Gibson and Paffenhöfer, 2000 for *D. gegenbauri*). Deibel, (1982b) found that ingestion rates for all zooids increased linearly with increasing zooid weight at similar rates, but the weight specific ingestion rates were independent of zooid weight. The mean daily ration for doliolids in that study was 132%. In our previous study, we determined that ingestion rates increased as food concentrations increased from 20 to 160 µg C/l of *Rho* and *T.w*, but decreased at an abnormally high concentration (Gibson and Paffenhöfer, 2000). The weight-specific ingestion rates ranged from 30 to 198%, and were highest at 160µg C/l, with an increase in increasing body weight, and lowest at 20µg C/l, here decreasing with increasing body weight.

The carbon content of *R. alata* (13.9 µg C/mm^3) was lower than that of *Rho* and *T.w* (160 µg and 80 µg C/mm^3 respectively), but in most cases *R. alata* represented the most carbon ingested among the three cells offered (Fig. 2A-C). The presence of the larger diatom affected the ingestion rates of the smaller co-occurring cells. The ingestion rates ranged from 6.0 to 30 µg C/zooid/d when gonozooids were offered *Rho* and *T.w.* only (Gibson and Paffenhöfer, 2000), however these rates were reduced from 5 to 15µg C/zooid/d when *R. alata* was included.

Although the presence of the larger volume *R. alata* cells inhibited the weight–specific ingestion rates of the small cells, the total carbon ingested did increase (Fig. 3). When only the small cells were offered at a concentration of 60 µg C/l in the previous study, the daily carbon ingested was 80 to 130% of the gonozooid's body carbon, as compared to 24-107% ingested from *Rho.* and *T.w.* in this study. As the total concentration of all cells increased from 80 to 160µg C/l, the daily ration ranged from 140 to 250% of body carbon ingested per day, when zooids were fed a mixed diet in this study; but was only 30 to 198% with just *Rho.* and *T.w.* at concentrations ranging from 20 to160 µg C/l. How will this affect growth and reproduction rates? Future experiments will be conducted to determine the effect of a mixed diet of *Rhizosolenia alata*, *Rhodomonas sp*., and *Thalassiosira weissflogii* with a higher carbon value than a diet of just *Rhodomonas sp*., and *Thalassiosira weissflogii*, on growth and reproduction rates.

SUMMARY

It was hypothesized that the presence of large diatoms would affect the feeding of gonozooids of *D. gegenbauri* on smaller co-occurring cells.

We found that:

- As *Dolioletta gegenbauri* gonozooids increased from 3.5 to 5.5 mm, their ability to ingest *R. alata* increased.
- The clearance rates of *R. alata*, *Rho*, *Tw*, and the *Rho+Tw* combination were not significantly different at the low concentration of *R. alata* for all gonozooid sizes.
- The small and medium sized gonozooids have the ability to ingest large and small cells at similar rates at all concentrations, while the high clearance rates of the larger gonozooids, at higher concentrations of *R. alata*, reduce the grazing pressure on smaller cells.
- The presence of *R. alata* inhibited the ingestion rates of smaller cells as compared to the previous study.
- The ingestion of these large diatoms increases the amount of carbon ingested per day, and may increase their rates of growth and reproduction.

Therefore, during upwelling events off the S. E. continental shelf when diatom abundances are increased, doliolids are able to ingest these cells at similar and higher rates than those of smaller co-occurring phytoplankton cells. Thus, reducing the abundance of a wide range of food particles available for co-occurring copepods and protozoan.

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REFERENCES

Crocker, K. M., Alldredge, A.L., and Steinberg, D. K. (1991) Feeding rates of the doliolid, *Dolioletta gegenbauri,* on diatoms and bacteria. *J. Plankton Res*., 13, 77 - 82.

Deibel, D. (1982b) Laboratory- measured grazing and ingestion rates of the salp, *Thalia democratica* Forskal and the doliolid, *Dolioletta gegenbauri* Uljanin (Tunicata, Thaliacea). *J. Plankton Res.*, 4, 189 - 201.

Deibel, D. (1985a) Clearance rates of the salp *Thalia democratica* fed on naturally occurring particles. *Marine Biology,* 86, 47 -54.

Deibel, D., and Paffenhofer, G, -A. (1988) Cinematographic analysis of the feeding mechanism of the pelagic tunicate *Doliolum nationalis*. *Bull. Mar. Sci.*, 43, 404-412.

Frost. B. W. (1972) The effects of size and concentration of food particles on the feeding behavior of the marine planktonic copepod *Calanus pacificus*. *Limnol. Oceanogr.y*, 17(6), 805-815.

Gibson, D.M. and Paffenhofer G.-A. (2000) Feeding and growth rates of the doliolid**,** *Dolioletta gegenbauri* Uljanin (Tunicata, Thaliacea*). J. Plankton Res*., 22, 1485-1500.

Harbison, G.R., McAlister, V.L. (1979) The filter-feeding rates and particle retention efficiencies of three species of Cyclosalpa (Tunicata, Thaliacea). *Limnol. Oceanogr*. 24(5), 875-892.

Mulling, M.M. (1983) *In situ* measurement of filtering rates of the salp Thalia democractica, on phytoplnakton and bacteria. *J. Plankton Res*., 5, 279-289.

Paffenhöfer, G.-A. and Knowles, S.C, (1978) Feeding of marine planktonic copepods on mixed phytoplankton. *Mar. Biol.*, 48, 143-152.

Paffenhöfer, G.-A. (1984) Food ingestion by the marine planktonic copepod *Paracalanus* in relation to abundance and size distribution of food. *Mar. Biol*., 80**,** 323-333.

Paffenhöfer, G. -A. and Lee, T. N. (1987) Summer upwelling on the Southeastern continental shelf of the U.S.A. during 1981. *Distribution and abundance of particulate matter*. *Prog. Oceanog.,* 19, 373-401.

Strathmann, R. A. (1967) Estimating the organic carbon content of phytoplankton from cell volume or plasma volume. *Limnol. Oceanog.*, 12, 411-418.

Tebeau, C. M., and Madin, L. P. (1994) Grazing rates for three life history stages of the doliolid *Dolioletta gegenbauri* Uljanin (Tunicata, Thaliacea). *J. Plankton Res.*, 16, 1075-1081.

Table I. Comparison of clearance rates of *D. gegenbauri gonozooids* and *T. democratica* aggregates at various particle sizes and Concentrations

Temperature (°C)	Species	Size (mm)	Food Type Concentration(ug C/l)	Clearance Rate (ml/d)	Reference
18-21	*T. democratica*	3 mm	*I. galbana* (4-5 um diam.)	53	Deibel 1982b
		4	+ *P. trochoideum* (16-18 um)	155	
		4.6	30 - 146 ug C/l	347	
	D. gegenbauri	2-18 ug C		10-335	
20-22	*T. democratica*	4.4-6.6	Natural particles 7 - 222 ug C/l	49-297	Deibel 1985
			(2-32 um diam) ESD		
12-14	*D. gegenbauri*	2.0-12.0	Bacteria <5 um dia	105	Crocker et. al 1991
			Nitzschia seriata > 100 um		
29	*D. gegenbauri*	5.0-10.0	Microspheres (2.5 um diam)	139	Tebeau & Madin 1994
20	*D. gegenbauri*	2.5-6.5	*Rhodomonas* (8um diam)	120-560	Gibson and Paffenhofer 2000
			T.weissfloggii (12 um) 60 ug C/l		
		3.5-5.5		113-870	Present study
			R. alata 21 ug C/l	320-870	
			46	100-680	
			100	94-360	

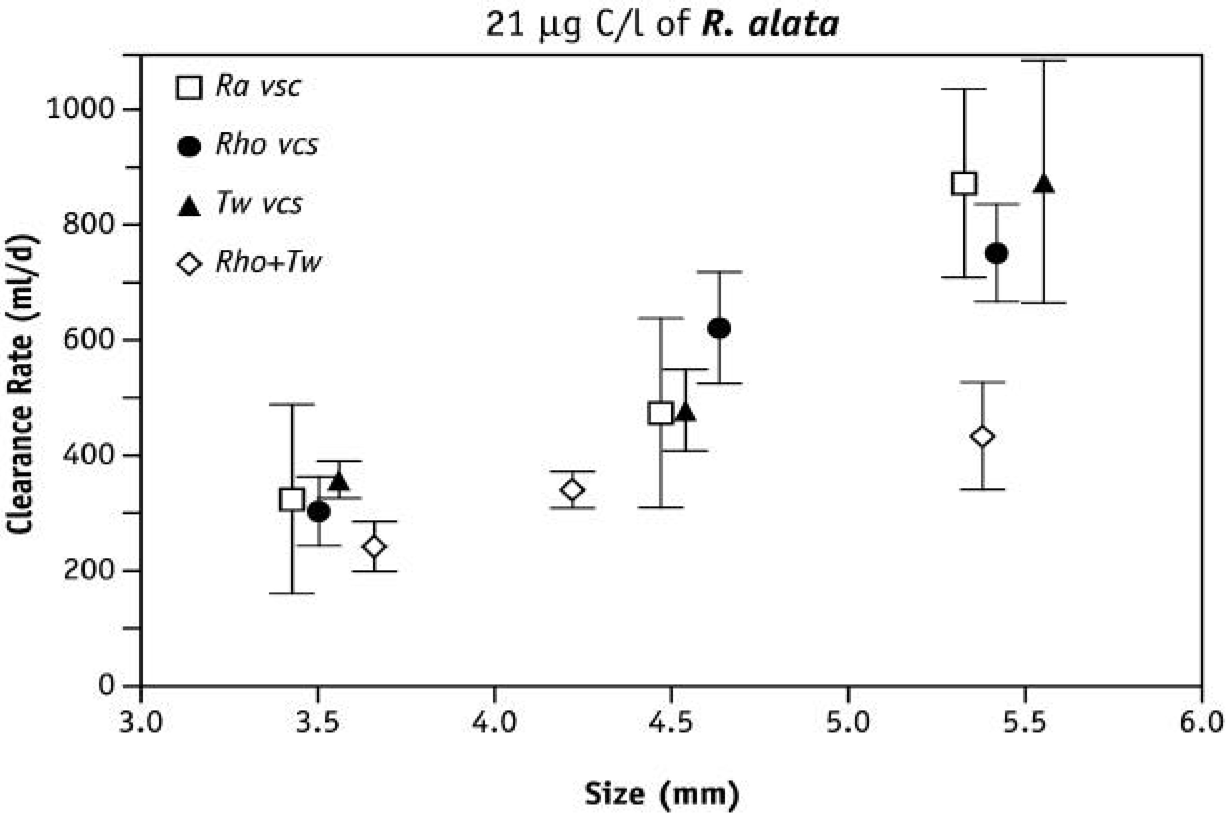

Figure 1A. Mean (± SE) clearance rates (ml • zooid^{-1} • d^{-1}) versus phytoplankton concentration (21 μg C • l^{-1}) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 3-13 observations.

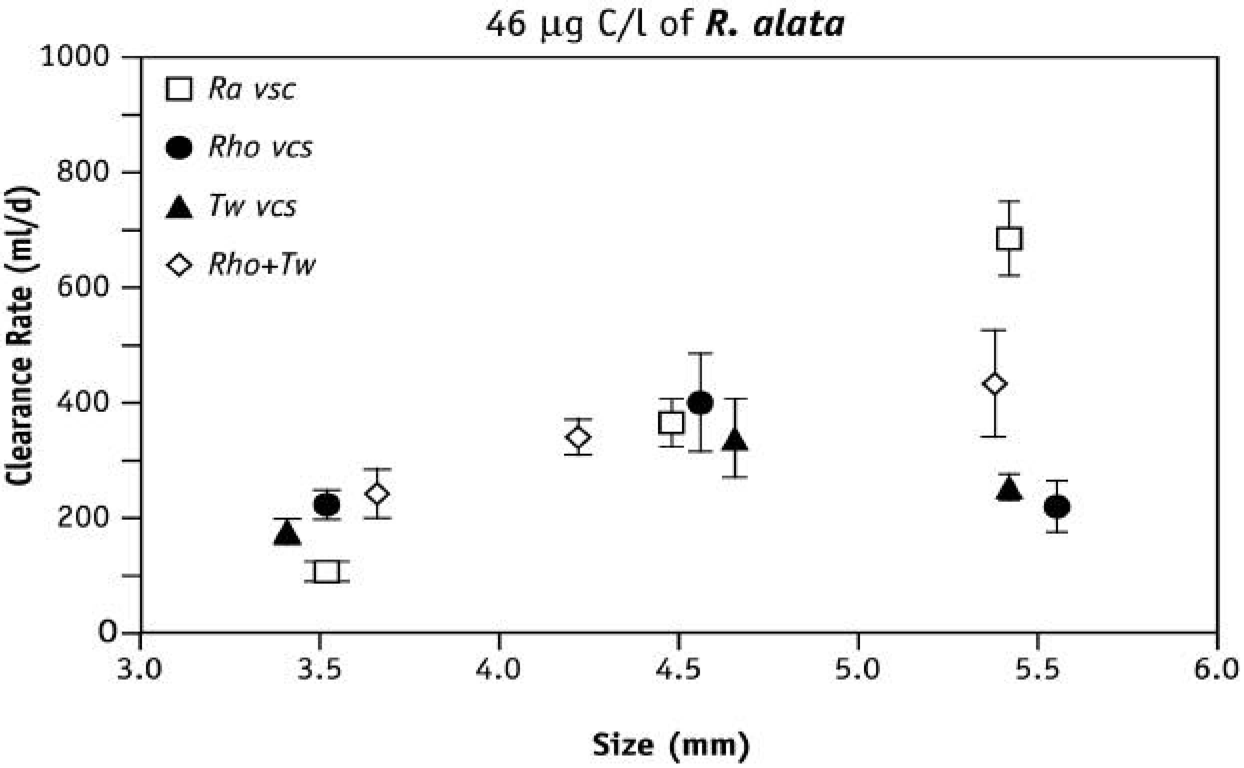

Figure 1B. Mean (± SE) clearance rates (ml • zooid^{-1} • d^{-1}) versus phytoplankton concentration (46 μg C • l^{-1}) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 3 - 13 observations.

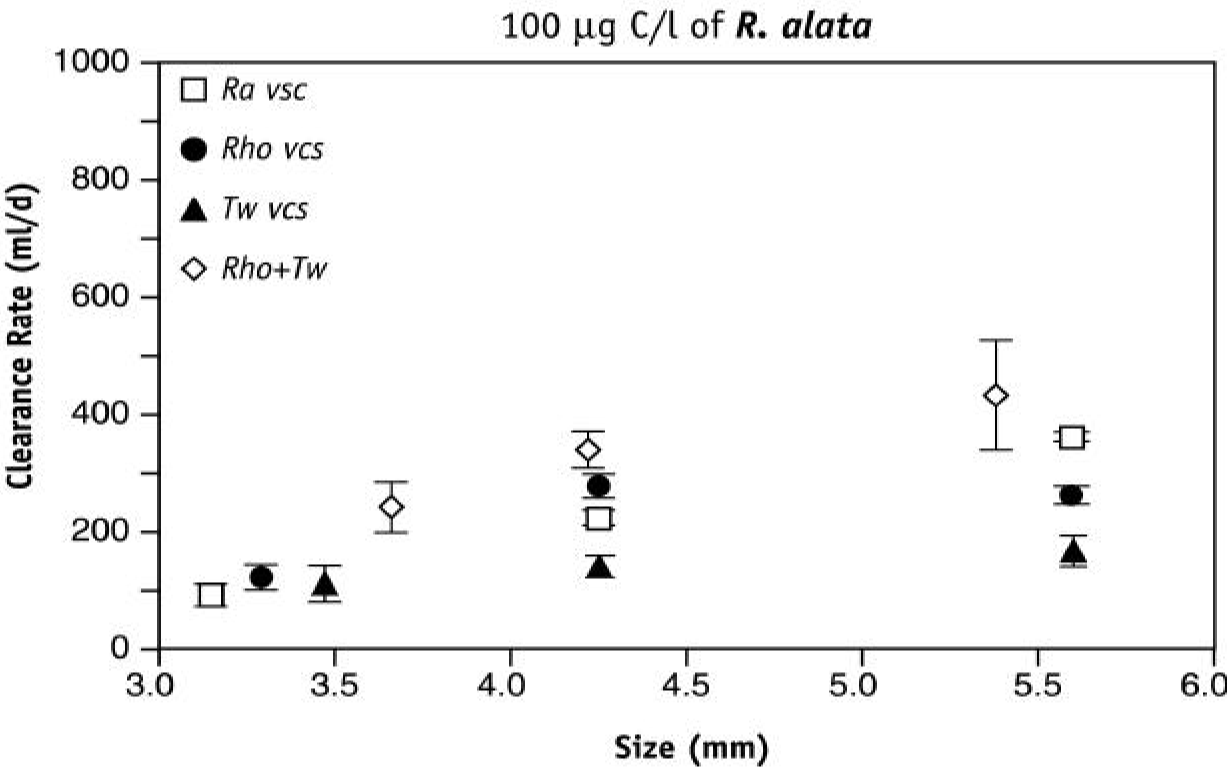

Figure 1C. Mean (± SE) clearance rates (ml • zooid^{-1} • d^{-1}) versus phytoplankton concentration (100 µg C • l^{-1}) for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 3 - 13 observations.

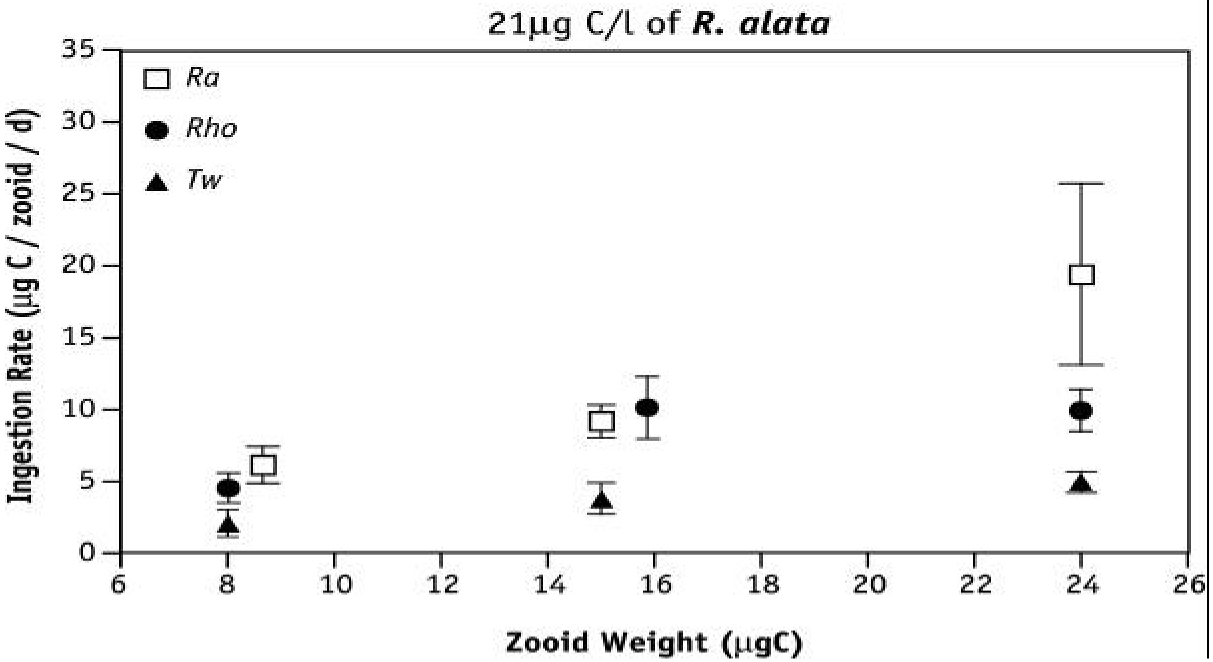

Figure 2A. Mean (+ SE) ingestion rates (µg C •zooid^{-1} • d^{-1}) at 21µg C/l of *R. alata* versus gonozooid weight for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 3 - 13 observations.

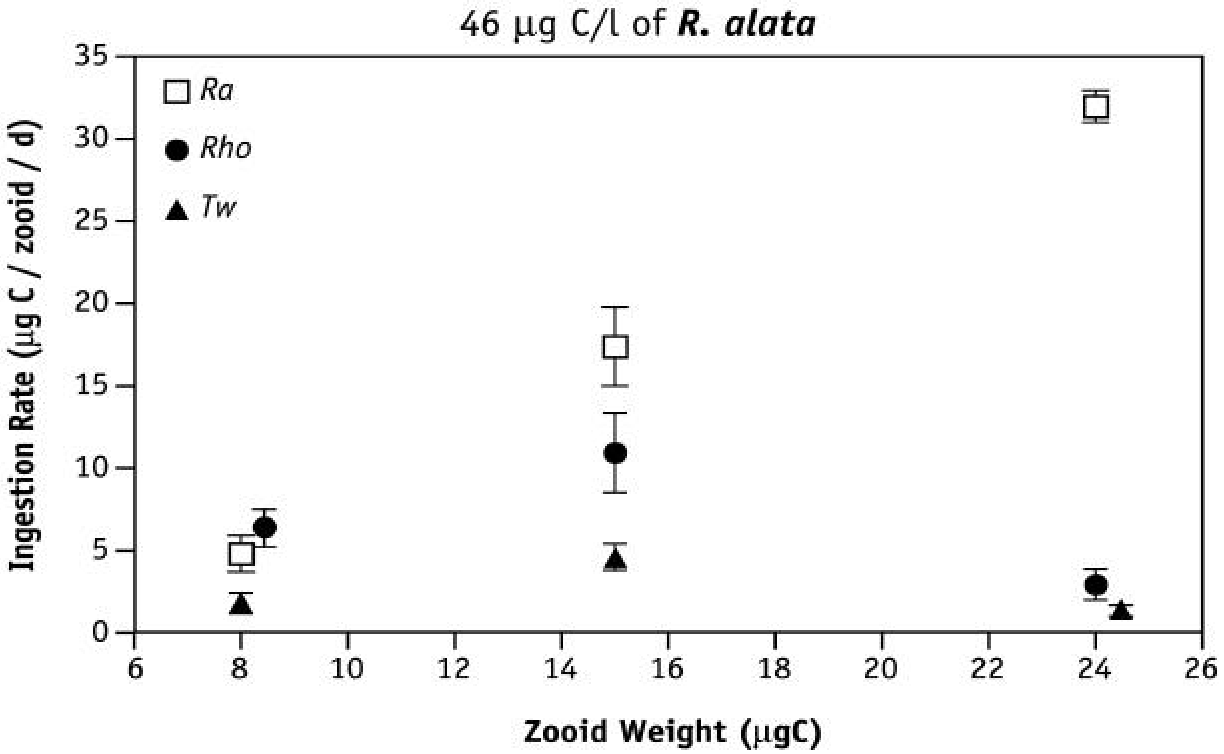

Figure 2B. Mean (± SE) ingestion rates (µg C •zooid^{-1} • d^{-1}) at 46µg C/l of *R. alata* versus gonozooid weight for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 3 - 13 observations.

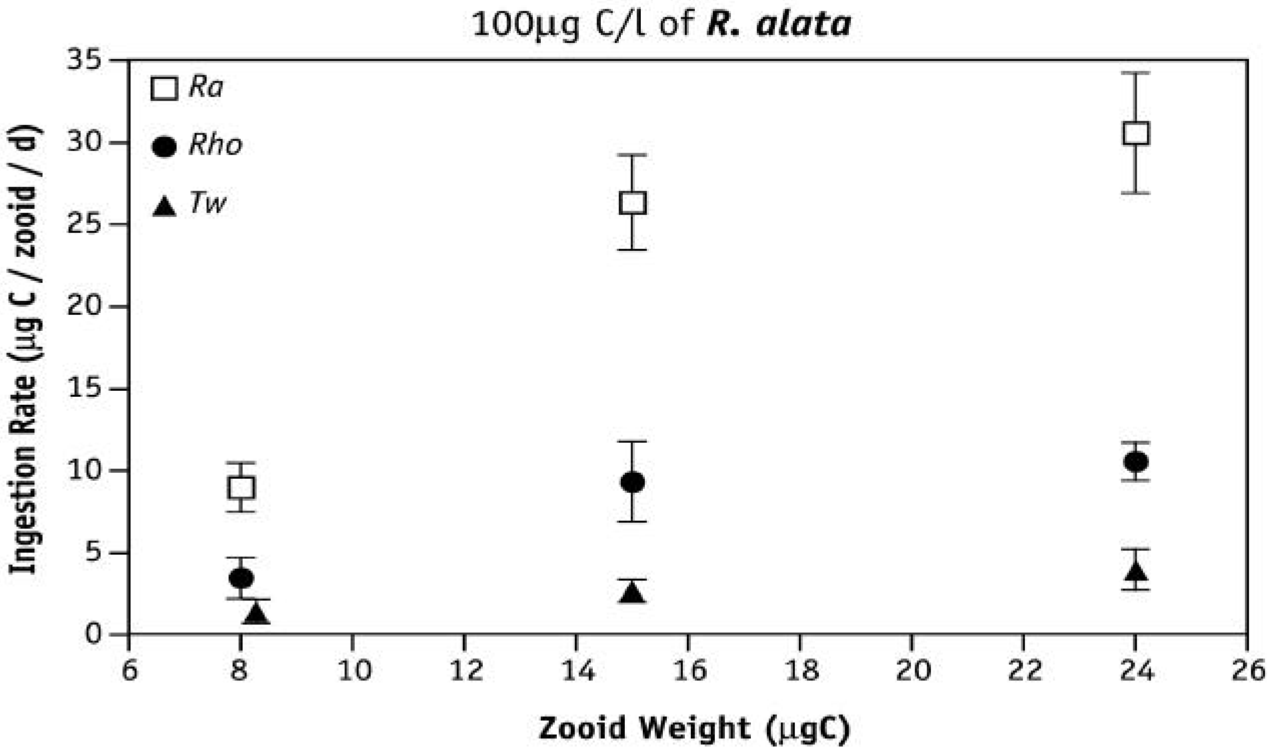

Figure 2C. Mean (± SE) ingestion rates (µg C •zooid^{-1} • d^{-1}) at 100µg C/l of *R. alata* versus gonozooid weight for three sizes of *Dolioletta gegenbauri* gonozooids. Each point represents 3 - 13 observations.

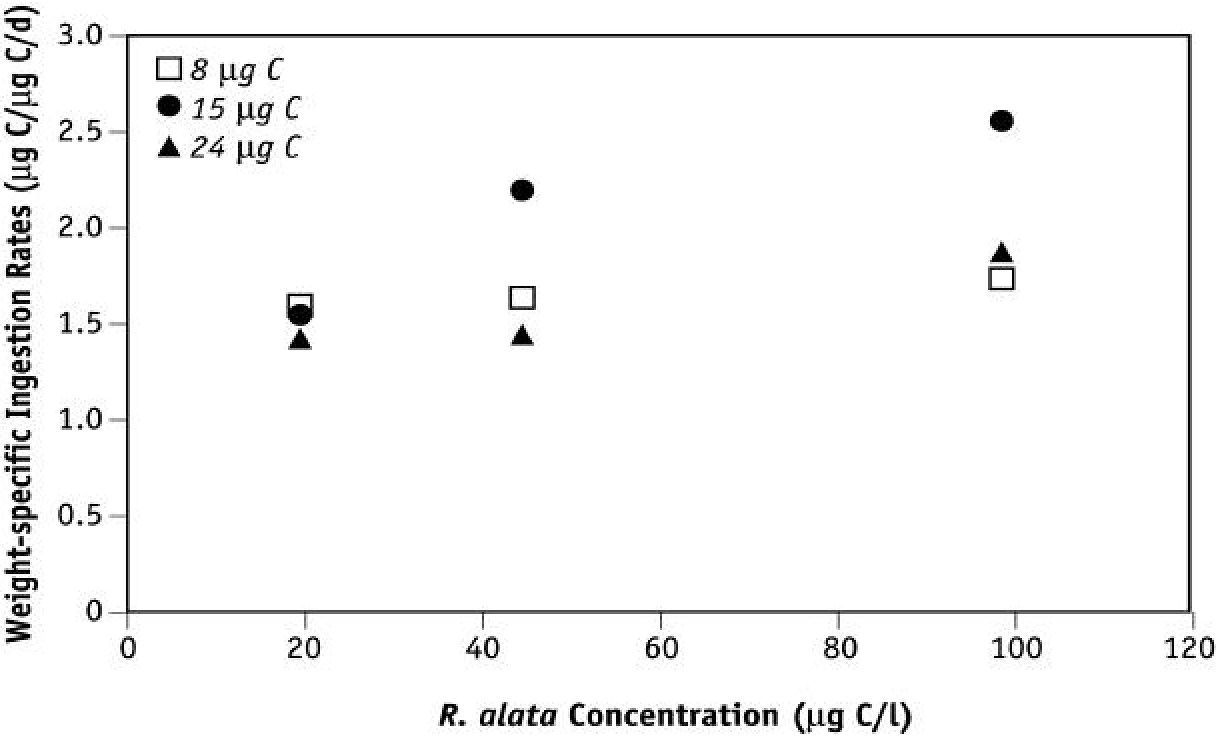

Figure 3. *Dolioletta gegenbauri*. Weight-specific ingestion rates of gonozooids of three different sizes in relation to *R. alata* concentration.

CHAPTER V

SUMMARY AND CONCLUSION

To understand the capabilities of an organism in nature, one must consider the effects of temperature and food concentration on rates of consumption, growth, and reproduction. The results from our studies allowed us to gain a better understanding of how these macrozooplankton function in the changing environment and how they may affect co-occurring planktonic organisms.

The overall goal of this study is to significantly enhance our knowledge on feeding, growth and reproduction of doliolids in order to be able to evaluate their contribution to the planktonic community as consumers and producers in comparison to copepods and protozoa.

Our objectives were to:

1. Quantify feeding and growth rates of gonozooids at environmental and experimental conditions.

Results from the feeding and growth experiments suggest that clearance rates are similar at concentrations from 20 to 60 µg C · l^{-1}, and decrease as the food concentrations increase to 160, and 390 µg C · l^{-1}. The ingestion rates increase over a range of phytoplankton concentrations from 20 to 160 µg C · l^{-1}, then decrease when abnormally high concentrations of 390 µg C l^{-1} are offered. Clearance and ingestion rates increase as temperature increases from 16.5 to 26.5º C. The exponential growth rates range from k = 0.2 - 0.7 with the lowest rates occurring at the highest food concentration. Growth rates increase with increasing temperature from K=0.1-0.3 at 16.5º C to 0.45-0.7 at 26.5º C. In each case, the small and medium sized zooids had higher growth rates than the larger gonozooids. These results suggest that doliolid feeding and growth rates are a function of environmental food concentrations and temperatures, and imply that they can be important consumers in a changing neritic environment.

2. Quantify rates of asexual and longevity of phorozooid reproduction at environmental and experimental conditions

Results from the reproduction experiments suggest that release rates increased at 20 °C as concentration increases from 7 to 160 µg C/l. Release rates remained similar as the temperature increased from 16.5 to 26.5 °C at a phytoplankton concentration of 60 µg C/l. Food concentration and temperature has an affect on phorozooid reproductive longevity, size of gonozooids released, and the amount of carbon released asexually. Doliolid reproduction rates are a function of environmental food concentrations and temperatures, and the results imply that doliolids can be important secondary producers in the neritic environment.

3. Quantify and compare feeding rates of gonozooids on a wide size range of food particles at environmental concentrations.

The results from these experiments suggest that as *Dolioletta gegenbauri* gonozooids increased from 3.5 to 5.5 mm, their ability to ingest *R. alata* increased. The small and medium sized gonozooids have the ability to ingest large and small cells at similar rates, while the larger gonozooid's clearance rates, at higher concentrations of *R. alata*, reduce the grazing pressure on smaller cells. The ingestion of these large diatoms increases the amount of carbon ingested per day, and may increase their rates of growth and reproduction.

CONCLUSION

Protozoans and copepods are considered the major consumers of primary production. They are found in lake, estuaries, and all parts of the world oceans, and can perceive and select their food. Thaliaceans, on the other hand, are restricted to coastal and neritic regions, and do not perceive nor select their food. However, it has been suggested that due to high feeding, growth, and reproduction rates, doliolids and salps may be able to control protozoa and calanoid copepods.

Dolioletta gegenbauri gonozooids have been found to impact an environment at a wide range of temperatures and food concentrations. We compared clearance and ingestion rates of copepods and doliolids to gain an understanding of how co-occurring species are maintained in the planktonic community. Trends for clearance and ingestion were the same for both species. As food concentrations increased, clearance rates decreased, and ingestion rates increased. An increase in temperature increased the rates of clearance, ingestion and growth for doliolids.

The presence of large diatoms decreased the grazing pressure on small cells, and increased the amount of carbon ingested by doliolids. Clearance rates and all cell sizes, were much higher than rates found by others investigating salps and doliolids, and higher than those of large co-occurring calanoid copepods. But, our growth rates were lower than salps and oikioplura, and higher than for calanoid copepods. There have been no assimilation efficiency experiments to explain how our high rates of ingestion support lower growth rates, or how other determined low ingestion rates support such high growth rates. More research is required to answer these important questions.

Knowledge of the impact on primary production by thaliaceans was limited due to the lack of investigation on the rates of reproduction over a wide range of food concentrations and temperatures. Our results were compared to the limited results on doliolids, and found that the releases of gonozooids by phorozooids were similar. We also compare our results with ones of co-occurring copepods species and found that daily and lifetime reproduction rates of doliolids surpassed those of copepods at similar or higher food concentrations.

With all results indicating doliolids having higher rates of ingestion, growth, and reproduction, why are copepods still the most dominate species in the planktonic community?

Copepods have several mechanisms to ensure survival in the planktonic community. Some species migrate to different regions in the water column during their life cycle to obtain food and avoid predators, while others lay dormant eggs to survive unfavorable conditions. There are no such mechanisms for doliolids. However, seed populations of dolioIds exist that are able to rapidly take advantage of phytoplankton blooms. Although doliolids have higher rates of clearance and ingestion, they are still unable to adjust their rates of feeding as copepods can, to maintain the % body carbon ingested at lower food concentrations. Doliolids have, increased ingestion rates with increasing temperature, which results in a higher % body carbon ingested. Due to the mechanics of thaliacean feeding, use of internal mucus nets, they are limited to coastal and oceanic regions. Copepods can increase their rates of ingestion at very low food concentrations to maintain similar amounts of carbon ingested per body weight by increasing their food perception. This may not affect doliolid populations as long as food levels remain near 20 µg C/l or higher. Phytoplankton concentrations and protzoplankton carbon can often be below 10µg C/l. If doliolid's clearance rates would remain near 500 ml/d at 8 µg C.l, they would only ingest 14% of their body carbon/day, wheras *E. pileatus* can ingest 27%. Thus allowing them to persist throughout the year.

Doliolids have two asexual stages that results in progeny close to the size of the adult and only takes four days to reproduce the next stage. This strategy can allow them to quickly colonize their environment, in two weeks, and with their high rates of feeding can out compete copepods during periods of upwelling. Copepods produce eggs at similar rates as doliolid phorozooid release rates of gonozooids, and have adapted many different strategies by brooding or releaseing eggs, and producing resting or diapause eggs to enhance their successfulness in so many parts of the world oceans.