

Impact of elevated temperature on the growth, survival, and trophic dynamics of winter flounder larvae: a mesocosm study

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Abstract: Winter flounder (*Pseudopleuronectes americanus*) is a dominant commercial fish in Narragansett Bay, Rhode Island, and yet factors controlling its recruitment remain unclear. An experiment was conducted with six 13-m³ land-based mesocosms (5 m deep) from February to April 1997 to address the impact of increased temperature (+3°C) on growth, survival, and trophic dynamics of winter flounder larvae. Objectives were to determine if warmer winter temperatures result in lower survival of winter flounder as a result of increased predator activity or if temperature-induced alterations in the food web result in greater food availability, perhaps leading to increased survival. Analyses of variance revealed significant ($P < 0.05$) or near-significant ($P < 0.10$) differences in phytoplankton and zooplankton abundance and biomass between warm and cool mesocosms. Winter flounder egg survival, percent hatch, time to hatch, and initial size were significantly greater in cool systems ($P < 0.05$). Mortality rates were lower in cool systems and significantly related to the abundance of active predators ($P < 0.05$). The cumulative impact of decreased survival of eggs and larvae in warm systems may partially explain the decline of winter flounder in Narragansett Bay, which has experienced elevated winter water temperatures in recent years.

Résumé : Bien que la Plie rouge (*Pseudopleuronectes americanus*) soit un poisson d'importance halieutique dans la baie de Narragansett au Rhode Island, les facteurs qui régissent son recrutement restent imprécis. Une expérience menée dans six mésocosmes de 13 m³ (5 m de profondeur), installés sur le sol, de février à avril 1997, a permis d'étudier l'effet d'une augmentation de température (+3°C) sur la croissance, la survie et la dynamique trophique des larves de la Plie rouge. Il s'agissait de savoir si une augmentation de température en hiver réduirait la survie de la plie à cause d'un surcroît de prédation ou si alors la température modifierait le réseau alimentaire et augmenterait la disponibilité de la nourriture, permettant peut-être ainsi une survie plus grande. Des analyses de variance révèlent des différences significatives ($P < 0,05$) ou presque significatives ($P < 0,10$) dans les densités et les biomasses du phytoplancton et du zooplancton entre les mésocosmes chauds et frais. La survie des oeufs de la Plie rouge, leur pourcentage d'éclosion, la durée de l'incubation et la taille initiale sont plus élevés dans les mésocosmes frais ($P < 0,05$). Les taux de mortalité sont plus bas dans les mésocosmes frais et ils sont en corrélation avec la densité de prédateurs actifs ($P < 0,05$). L'effet cumulé de la réduction de la survie des oeufs et des larves dans les mésocosmes chauds peut expliquer, au moins en partie, le déclin des populations de la Plie rouge dans la baie de Narragansett qui a connu des températures d'eau élevées en hiver depuis quelques années.

[Traduit par la Rédaction]

Introduction

Determining the causes of fluctuations in recruitment success of marine fishes remains a critical problem in fisheries biology. Evidence suggests that much of the variation in year-class strength is related to mortality of early life history stages (Houde 1987). The principal forces controlling recruitment success in larval fish are variations in food availability, particularly during the onset of first feeding, predation

by other organisms, and oceanographic conditions (Cowan and Houde 1990). The situation is further complicated by the potential interaction between any and all of these factors. Carbon dioxide production by our global industrial society has altered the earth's climate, with increased warming occurring in most regions of the Northern Hemisphere over the past century (Schuurmans 1995). In Narragansett Bay, Rhode Island, winter water temperatures significantly increased from 1959 to 1994 (Oviatt 1994). In a prior study, we determined that elevated winter temperature impacts the magnitude and fate of the winter-spring diatom bloom and thus may influence food availability to higher trophic levels (Keller et al. 1999a). Here, we extend our prior analysis to examine impacts on ichthyoplankton during a postbloom period. In addition to alteration in food availability, elevated water temperatures may lead to increased metabolic activity of potential predators followed by increased predation rates. This may be particularly important, since previous research examining the impact of nutrient enrichment on larval winter flounder (*Pseudopleuronectes*

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americanus) suggested a link between predation and survival (Klein-MacPhee et al. 1993). Here, we examine if elevated winter water temperature, due to global climate change, influences interactions between food availability and predation and leads to variation in survival of ichthyoplankton.

The life history and ecological niche of winter flounder are unique among the demersal fishes inhabiting east coast waters (Jeffries et al. 1989). Spawning generally occurs in shallow estuaries in winter and early spring at cold temperatures ranging from 1 to 10°C, with peak spawning at about 2–5°C (Pearcy 1962). The eggs are adhesive and demersal, with incubation lasting 5–31 days (Rogers 1976). Following hatching, the pelagic larvae are about 3 mm long but increase in size to about 7–9 mm over a 6- to 8-week period, at which time metamorphosis occurs (Laurence et al. 1979). Spawning at low temperatures appears to be a mechanism to “escape in time” from warm-water predators (Jeffries et al. 1989). Winter flounder success may thus be particularly sensitive to variation in temperature of the magnitude already observed in Narragansett Bay.

Historically, winter flounder has been the most abundant demersal fish in Rhode Island, contributing the bulk of the commercial and recreational bottom fishery ($>2 \times 10^6$ kg) in Narragansett Bay (Jeffries and Johnson 1974; Gibson 1998). Populations have tended to be somewhat cyclical throughout historical assessments, with adults known to fluctuate sevenfold within a decade (Jeffries and Terceiro 1985). Concerns are currently being expressed, since the population has dropped precipitously since 1979 and has remained low for the past 14 years (Gibson 1998). Inverse relationships between winter temperature and recruitment have been noted for winter flounder (Jeffries and Johnson 1974; Jeffries and Terceiro 1985; Northeast Utilities 1999). Jeffries and Terceiro (1985) have implicated climate warming and predator control as factors governing recruitment.

We hypothesized that warmer winter temperatures could result in elevated mortality of flounder larvae due to higher metabolic rate and increased activity of predators. In this paper, we test the hypothesized direct link between warmer temperatures and the growth and survival of winter flounder larvae and examine the relationship between temperature and the trophic components leading to larval fish. We describe the impact of increasing water temperature by 3°C relative to control systems. Control systems were maintained at a temperature 2°C less than the long-term (1977–1989) average for the parent system, Narragansett Bay. The study was conducted in six enclosed mesocosms of the Marine Ecosystem Research Laboratory (MERL) over a diatom-dominated post-winter–spring bloom period. We examined the effects of altered temperature on food availability (photosynthetic algae and zooplankton), abundance of active predators, and the growth and survival of winter flounder larvae.

Materials and methods

The MERL mesocosms used for the experiment are 13-m³ cylindrical enclosures with a 5-m water column scaled to the natural system, Narragansett Bay, in terms of physical, chemical, and biological characteristics. For this experiment, the six mesocosms were established as well-mixed systems with no exchange of sea-

water with Narragansett Bay after the start of the experiment. To induce natural levels of turbulence, mixing was applied via horizontal paddles located at 1-m depth intervals rotating with a rate of 4 rpm. The mixing schedule consisted of 2 h of mixing followed by 4 h of nonmixing, a scheme designed to simulate the semi-diurnal tidal currents in the Bay. Each mesocosm contained a 37-cm-deep sediment tray that was filled with lower Narragansett Bay sediment containing an intact (unmixed) benthic community. Sediments were collected with a 0.25-m² USNEL box core and remained undisturbed for 12 months prior to the start of the experiment. No attempt was made to manipulate the abundance of benthic organisms, which was intended to represent the range of natural variation seen at the sediment collection site. Previous studies have established that the mesocosms mimic the biological and chemical characteristics of the natural systems, including pelagic and benthic species composition, primary productivity, pelagic and benthic respiration, and nutrient concentrations and fluxes (see Sullivan and McManus 1986 and references therein).

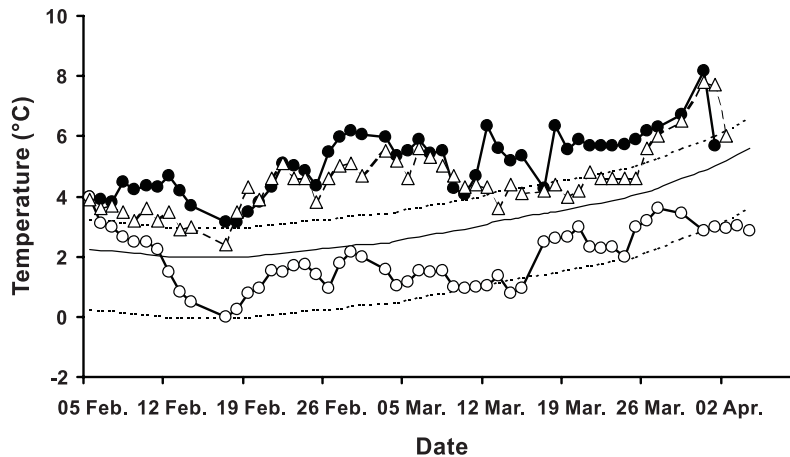
The experimental design consisted of two treatments using six outdoor mesocosms with three warm systems targeted for a temperature increase of +3°C relative to controls (also referred to below as cool). A time series based on the average monthly temperature in lower Narragansett Bay from 1977 through 1989 was used to set the temperature differential between warm and cool treatments (Fig. 1). Warm systems were heated 1°C relative to the long-term (1977–1989) Bay average, while controls were cooled 2°C below the average. Warm systems simulated the potential increase in winter water temperature expected to occur in the future, while cool systems represented typical winter temperature values experienced in the past (or current cold winters). Temperatures were controlled using polypropylene immersion type heat exchangers hung 2 m below the surface in each mesocosm. Triplicate treatments were established with natural plankton communities by filling the mesocosms with water from the adjacent Narragansett Bay. The experiment commenced on 5 February and ended on 4 April 1997.

Because winter flounder spawning tends to occur in the upper nutrient-enriched regions of Narragansett Bay, inorganic nutrients (NH₄Cl, KH₂PO₄, and Na₂SiO₃) were added daily to the six mesocosms. An N:P:Si molar ratio of 12.80:1.00:0.91 was used to mimic the ratio of nutrients in sewage effluents discharged into the Providence River (Oviatt et al. 1986). The loading level for nitrogen (2.31 mmol·m⁻³·day⁻¹), phosphate (0.18 mmol·m⁻³·day⁻¹), and silicate (0.165 mmol·m⁻³·day⁻¹) was four times the average annual loading (on an areal basis) to Narragansett Bay. The four times loading is within the range (two to eight times) occurring in the upper regions of the Bay where winter flounder larvae are most abundant (Oviatt et al. 1986; Keller et al. 1999b).

Temperature, chlorophyll *a* (Chl *a*), phytoplankton abundance, zooplankton abundance, and ichthyoplankton characteristics were measured in each of the six mesocosms. Water temperature (degrees Celsius) 10 cm below the surface was recorded daily. Replicate Chl *a* (micrograms per litre) was measured weekly from 10-mL aliquots of water subsampled from amber polyethylene bottles containing 2 L of water collected 1 m below the surface in each mesocosm during the morning mixing cycle. Samples were filtered through 25-mm Whatman GF/F glassfiber filters at 125-mmHg maximal vacuum. Two drops of 1% magnesium carbonate were added to each sample prior to filtration. Samples were extracted overnight in the freezer using 90% acetone and analyzed within 24 h on a Turner Designs model 10 field fluorometer (Lorenzen 1966).

Phytoplankton abundance (cells per millilitre) was measured weekly in each mesocosm from the amber bottles using standard techniques (Sournia 1978). Samples were preserved in Lugol's iodine solution and counted on an inverted microscope. Depending on phytoplankton density, either concentrated (10:1) or un-

Fig. 1. Ambient temperature model (solid line) used to set weekly temperature values for the study and the observed mean temperatures for cool treatments (open circles), warm treatments (solid circles), and Narragansett Bay (open triangles) over the experimental period (5 February – 4 April 1997). The ambient model was based on the mean monthly temperatures in Narragansett Bay in the lower West Passage from 1977 through 1989. Warm tanks were targeted for a +1°C (upper broken line) increase above ambient and cool tanks for a -2°C (lower broken line) decrease below the model as shown in the figure.



concentrated samples were counted by pipetting a 2-mL subsample into a 5-mL settling chamber and adding 3 mL of artificial seawater. Samples were settled for 20 h and a known area of the chamber counted (one to two diameters, half chamber, whole chamber) until at least 1000 cells were counted.

Zooplankton samples were collected weekly by pumping 50–100 L of water at a constant rate from each of five depth intervals (0.1, 0.5, 1.0, 2.5, and 4.5 m). Pumped samples were integrated over each depth interval (e.g., 1.0 m represents 0.5–1 m) by raising the sample hose at a constant rate. Zooplankton larger than 44 μm were filtered from the water samples, preserved in 10% formalin, staged, identified, and enumerated. Zooplankton were identified to species or genus level for adult and copepodite stages and to order or class for nauplii or other invertebrate larvae.

Winter flounder eggs and larvae were obtained from broodstock collected in Narragansett Bay primarily by otter trawl in early December 1996. Adult fish were sorted by sex and maintained in separate outdoor tanks supplied with running seawater at ambient temperature and salinity until ripe. Fertilized eggs were obtained by mechanically stripping two ripe females and three ripe males using the methods of Klein-MacPhee et al. (1982). Approximately equal numbers of newly spawned eggs (~5000) were placed in replicate egg baskets suspended at 1 m depth in each mesocosm at the start of the experiment (5 February) at temperatures of 3.9–4.0°C. Temperatures were gradually raised in the warm treatments and lowered in the cool treatments to achieve the desired 3°C temperature differential within the first week of the experiment. The cylindrical egg baskets (500 mL volume, 10 cm in diameter) were constructed of plastic with mesh (250 μm) caps and bottoms. Baskets were designed to allow incubation with good water circulation at experimental temperatures. Eggs were suspended at 1 m depth, since winter flounder eggs are demersal and occur naturally at this depth (Crawford 1990). Egg density in the baskets was set by estimating the total number of eggs required for weekly sampling (prior to hatching) plus the number of larvae needed to stock the mesocosms (after accounting for egg mortality). Egg density has not been estimated in the field, but winter flounder produce approximately 500 000 demersal eggs at spawning. Weekly subsamples of about 200 eggs were removed via pipetting (5 February – 7 March) from each egg basket and examined microscopically to determine the number of live versus dead eggs. Percent egg mortality was calculated directly from the ratio of dead to total eggs counted.

To determine percent hatch, a subsample of 100 eggs was counted and placed in replicate 4-L plastic containers with a 150- μm -mesh bottom in each mesocosm on 5 February. Eggs in these containers were incubated at depths of 0.2 m, which is shallower than the norm but allowed a rapid visually determination of when hatching occurred. Upon hatching, contents of each container were retrieved and the number of larvae counted. Percent hatch for each mesocosm was determined directly as the ratio of live larvae to eggs incubated.

After hatching, the egg baskets in each mesocosm were retrieved and newly hatched larvae were used to stock the system in which they were suspended. Larvae from replicate egg baskets were counted as they were gently poured into 4-L polycarbonate containers, combined in a 1:1 ratio, and released into the mesocosm in which they hatched. A total of 2000 larvae were added to each mesocosm. Stocking density for each mesocosm was ~150 larvae·m⁻³, at the upper range of densities (100–150·m⁻³) for winter flounder larvae occurring naturally (Pearcy 1962). Because winter flounder are currently at low population levels, stocking density was based on older larval density data, which are more in line with the carrying capacity of the system. Additionally, density of newly hatched larvae in the field is rarely reported since these small larvae tend to be undersampled using standard-mesh (505 μm) ichthyoplankton nets (Northeast Utilities 1999). A subsample of 30 larvae from each mesocosm was measured at hatching. Initial lengths (standard length, millimetres) were obtained by anesthetizing larvae with tricane methanesulfonate (MS 222) (to prevent curling) and measuring them alive under a dissecting microscope.

Fish larvae were collected weekly in two replicate samples using a 0.5-m-diameter, 202- μm -mesh plankton net equipped with a TSK flowmeter. Replicate tows were made with the mixers off by lowering the net to the bottom of the mesocosm and pulling it steadily to the surface. Flowmeter readings were recorded before and after each tow. Water and organisms were briefly stored in a cold room in 4-L plastic containers until the larval fish could be removed, anesthetized as above, and measured alive. The number of larvae caught per tow was used to estimate larval abundance in the mesocosm over the course of the experiment.

When the experiment was terminated, a circular (1.75-m-diameter) 333- μm -mesh net was lowered to the bottom of each mesocosm. The water column in each mesocosm was then drained through a 202- μm -mesh plankton net via a valve located 0.5 m from the bottom. Any larvae remaining in the mesocosm after

draining were collected by raising the circular net through the 0.5 m of water still in the mesocosm. The total number of larvae surviving at the end of the experiment in each mesocosm was obtained directly by counting the larvae collected in both the overflow net and the bottom net. The total count of larvae obtained when the mesocosm was completely drained at the end of the experiment was then compared with the final net tow estimate (just prior to draining) to determine capture efficiency. Data were not corrected where the capture efficiency were low because capture efficiency varies with age (Klein-MacPhee et al. 1993).

Larval daily instantaneous growth rates (assuming exponential growth) were calculated via regression using the equation

$$(1) \quad G = (\log_e SL_t - \log_e SL_0)/t$$

with G the daily growth rate, SL_t the larval standard length (millimetres) at time t , SL_0 the larval standard length at $t = 0$, and t the time interval (days).

Average daily instantaneous mortality rates were calculated as

$$(2) \quad Z = (\log_e N_0 - \log_e N_t)/t$$

with Z the mortality coefficient (per day), N_t the number of winter flounder surviving at time t , N_0 the number stocked at the beginning of the study (time $t = 0$), and t the time (days). Following Laurence (1974), N_t was corrected for potential survivors removed by sampling using the formula

$$(3) \quad N_{t'} = N_t + R \sum_{K=1}^{w-1} e^{-7KZ}$$

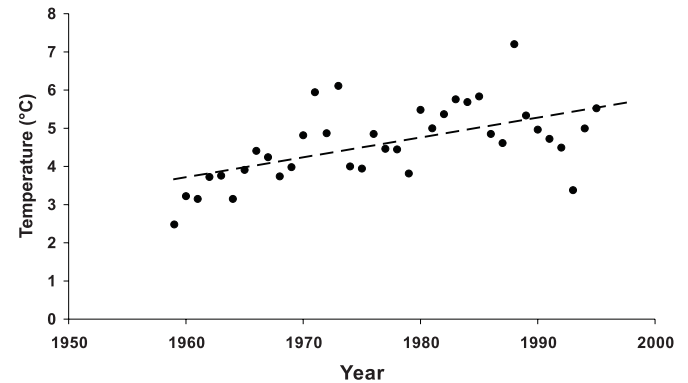
with R the number of individuals removed each week by sampling, w the number of weeks of sampling, 7 the number of days in 1 week (adjusted if sample intervals were unequal), and Z the average daily instantaneous mortality coefficient. The initial Z value in eq. 3 is the maximum mortality rate from eq. 2 based on no survivors from samples taken and was used to calculate iteratively $N_{t'}$. The $N_{t'}$ was then substituted for N_t in eq. 2 to calculate an unbiased estimate of Z .

On 21 March 1997, the egg basket in a cool system (tank 5) was spilled, releasing an unknown number of eggs and larvae into the mesocosm. Yolk-sac larvae were collected on sampling. No yolk-sac larvae were observed on the date prior to the spill. Growth and mortality rates for this system were calculated from data collected up to the time of the spill.

The abundance of benthic predators was censused by diver survey prior to the start and near the end of the study. Numbers were confirmed following draining of the mesocosms when the experiment was terminated. Pelagic predators were determined from the net samples (ring net and overflow net) collected when the entire system was drained at the completion of the study.

We statistically assessed the effect of elevated temperature on variables measured over time throughout the entire experimental period. We used a nested analysis of variance (ANOVA), which is equivalent to a repeated-measures analysis (Winer 1971). For this design, replicate mesocosms were nested within treatments and considered random. In testing for main effects (treatments controlled as part of experimental design), sample date was considered a repeated factor and the degrees of freedom of error terms were not artificially inflated by frequent sampling. In those cases where there was a significant time-by-treatment interaction, we used a nested ANOVA on each sample date where replicate mesocosms were again considered random. Abundance data were transformed ($\ln(x + 1)$) to reduce heterogeneity of variance prior to statistical analysis, and t tests were used to determine significant differences between larval winter flounder growth and mortality rates between treatments.

Fig. 2. Increase in temperature during the peak winter flounder larval period (February–April) in Narragansett Bay from 1959 to 1998. Data were collected at hourly intervals using a buoy moored in the East Passage of the Bay by the National Oceanic and Atmospheric Administration. Winter temperature was calculated as the mean of the monthly temperatures. The regression equation ($Y = 0.053X - 100$) indicates an average increase of $0.053^\circ\text{C}\cdot\text{year}^{-1}$ for a total increase of 2.1°C over the 40-year period.



Results

Water temperature in Narragansett Bay during the winter flounder larval period (February–April) has increased significantly ($P < 0.05$) over the past 40 years ($+0.053^\circ\text{C}\cdot\text{year}^{-1}$) (Fig. 2). For the current experiment, water temperature in the warm treatments ranged from a low of 2.9°C in February to a high of 8.3°C in late March, with an average of 5.11°C (± 0.14 SE) over the experimental period (Fig. 1). The cool systems increased from -0.1 to 3.7°C over the same period, with a mean temperature of 1.86°C (± 0.13 SE). Since temperature did not vary significantly within treatments ($P > 0.05$), daily means are shown by treatment (Fig. 1). These values are plotted relative to the weekly model temperatures: warm ($+1^\circ\text{C}$), mean, and cool (-2°C). The actual temperature difference between treatments was 3.25°C , close to the targeted differential of 3°C . Water temperature in Narragansett Bay over the same period (Fig. 1) was warm relative to long-term data, with an average temperature of 4.4°C , but on average 0.7°C cooler than the warm treatments.

Both phytoplankton abundance ($\sim 10\,000$ cells $\cdot\text{mL}^{-1}$) and biomass as Chl *a* (~ 15 $\mu\text{g}\cdot\text{L}^{-1}$) were relatively high at the start of the experiment, suggesting a minor bloom in the Bay at the time of the fill (Fig. 3). Abundance (Fig. 3A) and biomass (Fig. 3B) increased gradually in both treatments over the first 3 weeks of the experiment, with both measurements being somewhat higher in cool mesocosms. From late February through late March, total cell counts were generally higher in the warm treatments (ANOVA, $F = 1.9$, $P < 0.09$, near significance), while biomass was significantly greater in these systems (ANOVA, $F = 4.5$, $P < 0.001$) (Table 1). At the end of the experimental period, abundance and biomass were significantly greater in the cool tanks.

In general, *Detonula confervacea* was the dominant diatom in both warm (Fig. 3C) and cool (Fig. 3D) systems during the initial bloom period and was not significantly different between treatments (Table 1). Following the initial

Fig. 3. (A) Total phytoplankton cell counts and (B) Chl *a* shown as the mean (± 1 SE) by treatment from 6 February to 4 April 1997. Warm tanks are represented by solid circles and cool tanks by open circles. Phytoplankton abundance for the dominant species is shown for (C) a representative warm system and (D) a representative cool system from 6 February to 4 April 1997.

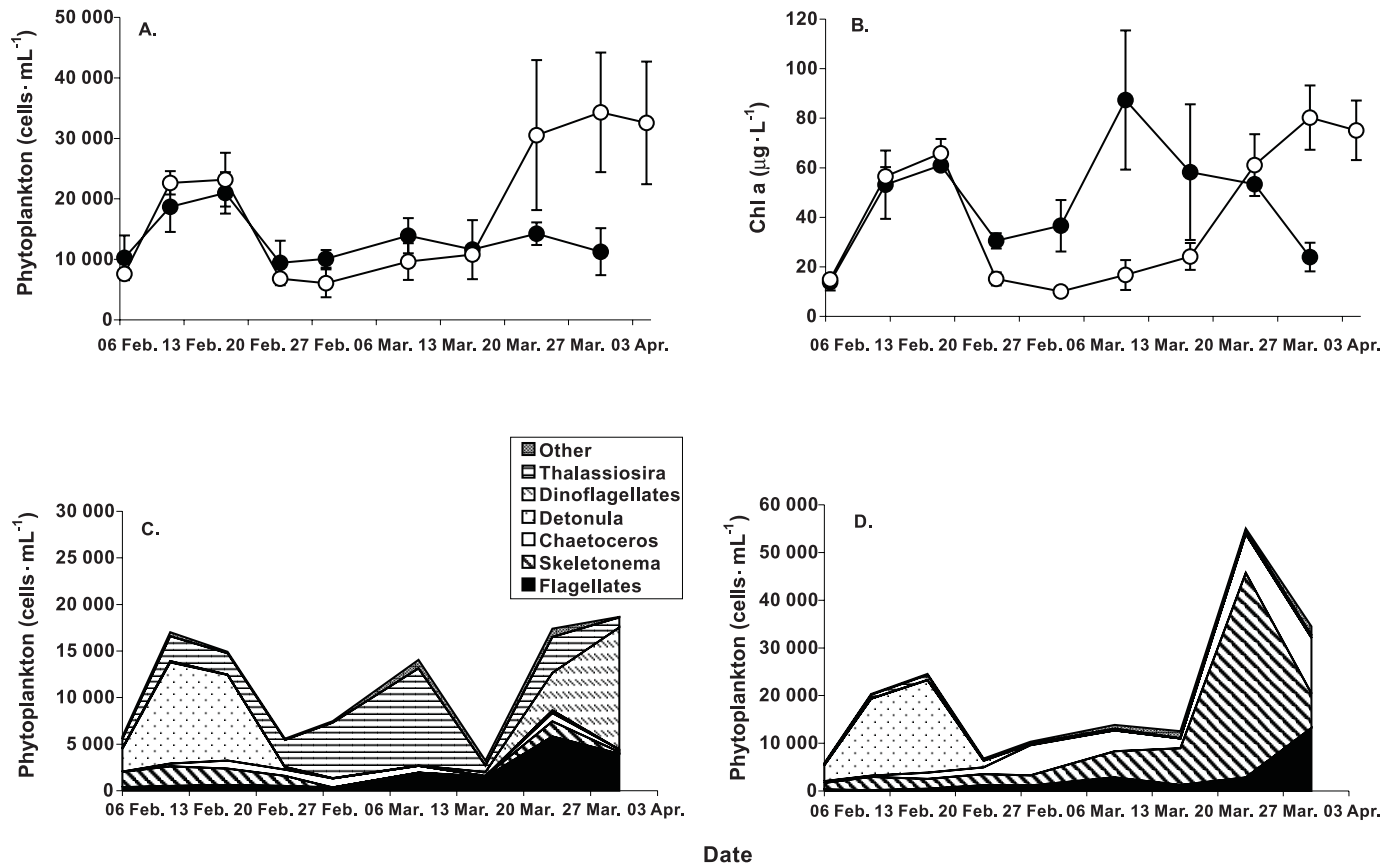


Table 1. Results of nested ANOVAs (equivalent to repeated measures analyses) showing main effects and time-by-treatment interactions of experimental manipulations (warm versus cool mesocosms) on phytoplankton and zooplankton.

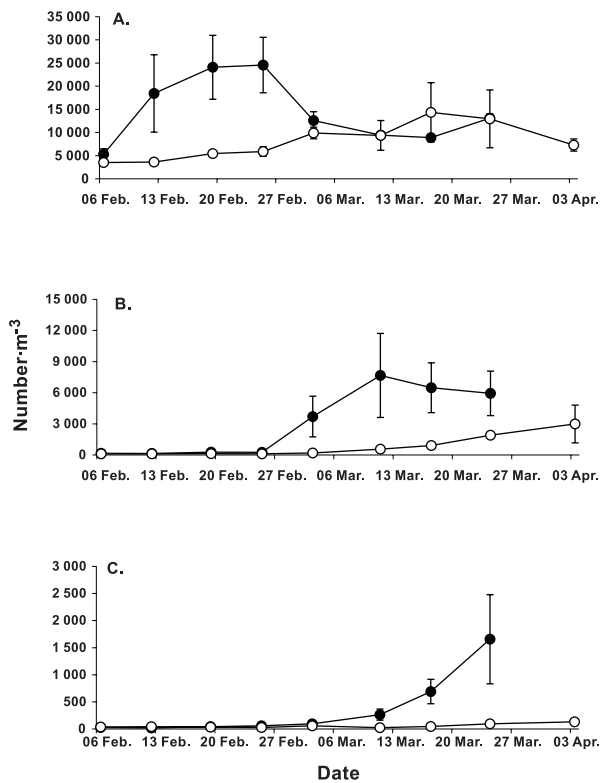
Variable	Treatment effects		Time-by-treatment interactions	
	F	P	F	P
Phytoplankton (cells·mL ⁻¹)				
<i>Skeletonema</i>	23.4	0.008	4.2	0.002
<i>Chaetoceros</i>	21.8	0.009	6.3	0.0001
<i>Thalassiosira</i>	22.1	0.009	2.1	0.05
<i>Detonula</i>	0.72	0.45	0.4	ns
Flagellates	0.02	0.91	3.3	0.007
Dinoflagellates	0.01	0.95	2.4	0.04
Total	2.6	0.18	1.9	0.09
Chl <i>a</i> (µg·L ⁻¹)	5.8	0.07	2.4	0.04
Zooplankton (number·m ⁻³)				
Nauplii	4.8	0.09	6.5	0.0001
Copepodites	7.2	0.05	5.4	0.0004
Adults	6.9	0.06	2.6	0.03
Total	5.7	0.07	2.9	0.02

Note: Abundance data were natural logarithmically transformed prior to analysis ($n = 52$). ns, not significant.

3-week period, the species composition between treatments began to diverge. The warm systems (Fig. 3C) were characterized by a significant increase in *Thalassiosira* spp. that did not occur in the cool systems (Fig. 3D). *Skeletonema costatum* and *Chaetoceros* spp. were the dominant species present in the cool systems during this period (Fig. 3D). *Skeletonema* and *Chaetoceros* spp. continued to dominate the cool systems during the final weeks of the study. While *Skeletonema* was present in warm systems during these last few weeks, the phytoplankton population was primarily composed of flagellates, dinoflagellates, and *Thalassiosira* spp. Flagellates increased in abundance in both treatments towards the end of the study (Figs. 3C and 3D).

Significant ($P < 0.05$) and near-significant ($P < 0.10$) treatment effects were noted for biomass as Chl *a*, total phytoplankton abundance, and abundance of *S. costatum*, *Chaetoceros* spp., and *Thalassiosira* spp. over the experimental period (Table 1). Phytoplankton biomass and cell counts were elevated in the warm treatment relative to the cool until the final week of the study. *Skeletonema* and *Chaetoceros* spp. were more abundant in cool systems, while *Thalassiosira* spp. were more abundant in warm systems. The levels of dinoflagellates and flagellates were not significantly different throughout the experimental period but exhibited significant time-by-treatment interactions (Table 1). ANOVA by sample date indicated significantly more dinoflagellates and flagellates in warm systems from late February

Fig. 4. Copepod abundance shown as the treatment means (± 1 SE) by stage, (A) nauplii, (B) copepodite, and (C) adult, from 6 February to 4 April 1997. Warm tanks are represented by solid circles and cool tanks by open circles.



through March. The levels of *D. confervacea* were not significantly different between treatments throughout the study (Table 1).

Mean abundance (\pm SE) of copepods (by stage) for the triplicate control and treatment mesocosms over the experimental period is displayed in Fig. 4. Mean abundance of nauplii ($<5000\text{-m}^{-3}$) at the start of the experiment was low, reflecting the abundance of nauplii in Narragansett Bay at the time of the fill, but increased substantially in warm systems ($\sim 23\,000\text{-m}^{-3}$) relative to controls ($\sim 10\,000\text{-m}^{-3}$) during February. From 3 March 1997 through the remainder of the experiment, the abundance of copepod nauplii was similar in both treatments. At the start of the experiment, the abundance of copepodites and adult copepods was also low in all mesocosms ($<150\text{-m}^{-3}$). Copepodite numbers increased somewhat earlier (5 March) and to a greater extent in the warm systems relative to cool systems (Fig. 4). A similar pattern was observed for adult copepods. In the warm systems, abundance of adults increased markedly from 11 March through the end of the study. In the cool systems, very slight increases were observed from 11 March to 3 April 1997.

Copepod nauplii, copepodites, adult copepods, and total zooplankton levels were significantly (ANOVA, $P < 0.05$) or close to significantly (ANOVA, $P < 0.10$) greater in the warm versus the cool treatments over the experimental period (Table 1). When differences were not significant over the entire period, there were significant time-by-treatment interactions that, when examined by sample date, indicated

significant differences in copepod nauplii and total zooplankton during the initial experimental period (12–25 February 1997). Abundance of copepodites varied significantly between treatments from 25 February – 24 March, while adult values were significantly greater in warm systems from 17 March to the end of the study (Table 1). The copepod community (adults and copepodites) was dominated (58–84%) by *Acartia hudsonica* in all systems. Both *A. hudsonica* and *Centropages hamatus* were significantly more abundant in the warm treatments (ANOVA, $P < 0.05$). The remainder of the zooplankton community, composed primarily of *Temora longicornis*, *Eurytemora affinis*, *Oithona* spp., harpacticoid copepods, barnacle nauplii, and polychaete larvae, showed no significant differences between treatments (ANOVA, $P > 0.05$).

Both phytoplankton (as Chl *a*, micrograms per litre) and zooplankton are important food sources for larval winter flounder (Laurence 1977; Klein-MacPhee et al. 1993). In addition to diatoms and dinoflagellates, winter flounder larvae feed primarily on copepod nauplii, calanoid and harpacticoid copepods, polychaete larvae, and invertebrate eggs. Total food availability (both phytoplankton (Fig. 3) and zooplankton (Fig. 4)) was generally higher in the warm systems relative to the cool over most of the experimental period. Mean zooplankton abundance (number per cubic metre) increased with mean phytoplankton biomass (Chl *a*, micrograms per litre) in the warm and the cool systems and was positively related (ANOVA, $F = 4.0$, $P = 0.1$, near significance) to temperature (Fig. 5). Our results indicate that food availability was not a primary factor controlling larval mortality, since mortality was higher in the warm systems (see below) where more food was present.

Hatching of winter flounder eggs in the warm systems occurred on 25 February after a 20-day incubation period at an average temperature of 4.1°C (± 0.1 SE). Hatching in cool systems occurred on 7 March following a 30-day incubation period at an average temperature of 1.6°C (± 0.2 SE). Egg mortality (percent) was not significantly different between treatments during the first week of the study but increased significantly (ANOVA, $F = 7.62$, $P < 0.05$) in the warm systems by 19 February and remained higher until hatching (Fig. 6). Percent hatch for the warm treatments ranged from 64.6 to 81.8%, while the range in cool systems was 91.3–94% (Fig. 7). Percent hatch in the cool systems was significantly greater than in the warm (t test, $P < 0.05$).

The size at hatching (mean standard length \pm SE) in the cool systems (3.2 ± 0.01 mm) was significantly larger (t test, $P < 0.05$) than the size at hatching in warm mesocosms (3.0 ± 0.01 mm). The mean and range in larval standard length gradually increased over the experimental period in each mesocosm (Fig. 8). In general, the exponential model for growth (eq. 1) gave a good fit to growth in standard length of larval winter flounder over time as indicated by low standard errors for the estimated growth rates (Table 2). Instantaneous growth rates in the warm systems ranged from 0.016 to 0.018-day^{-1} and were higher than values in the cool systems (range 0.011 – 0.013-day^{-1}) (Table 2). Growth rates were compared with analysis of covariance (ANCOVA) and tested for heterogeneity of slopes (since growth rates are the slopes of regression equations) between treatments. Instantaneous daily growth rates of winter flounder larvae were

Fig. 5. Empirical relationship ($Y = 609.3X - 8389$, $r^2 = 0.50$) between mean zooplankton abundance and mean phytoplankton biomass (Chl *a*) in the warm (W) and cool (C) treatments.

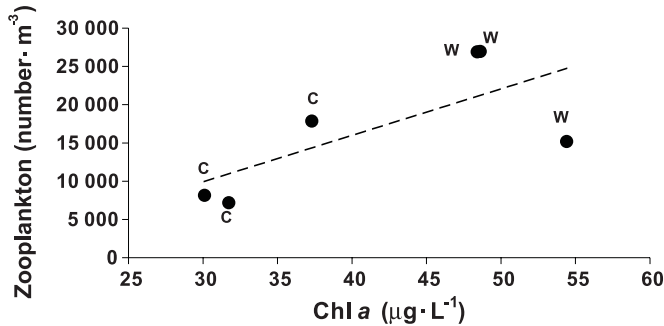
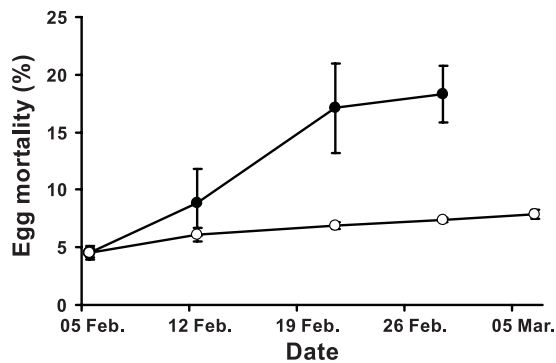


Fig. 6. Percent mortality (± 1 SE) for winter flounder eggs over the incubation period for warm (solid circles) and cool treatments (open circles).

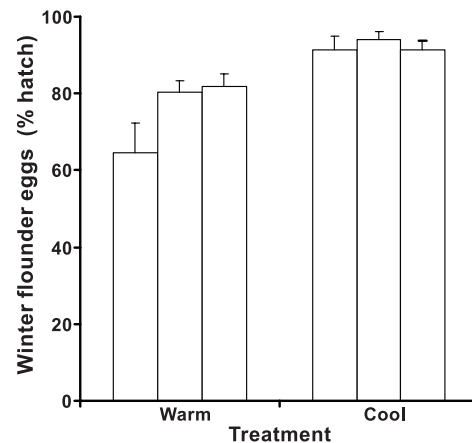


significantly greater in the warm versus the cool systems (ANCOVA, $F = 14.54$, $P = 0.0001$). Although initially smaller, the larvae in the warm systems grew more rapidly and reached a larger size than larvae in the cool systems by the end of the experiment.

Following stocking, winter flounder populations in both treatments were characterized by an initial period of rapid decline followed by a period of decreasing mortality (Fig. 9). Recall that with the exception of the final sample, abundance in each mesocosm was estimated by sampling with a plankton net. When the tanks were drained at the end of the experiment and the entire population censused in each system, the capture efficiency for the plankton nets was 55–98%. The final abundance estimate for calculating mortality rate in each mesocosm was based on the total population count rather than the co-occurring final estimate from sampling with the plankton net. Instantaneous daily mortality rates of winter flounder larvae (eq. 2) were higher in the warm enclosures (Table 3). ANCOVA indicated that differences in mortality rates were near significance (ANCOVA, $P < 0.07$) in the warm versus the cool systems.

Two potentially important benthic predators on winter flounder larvae were present in the mesocosms throughout the study (Table 4), the sand shrimp (*Crangon septemspinosa*) and the mud anemone (*Cerianthopsis americana*) (Holohan 1993; Thornton-Whitehouse 1994). Although *Crangon* were present in the cool tanks, they were inactive and buried in the sediment. In the warm systems, *Crangon*

Fig. 7. Percent hatch (± 1 SE) for winter flounder eggs in the three warm and three cool treatments based on replicate samples of 100 eggs placed in 4-L plastic containers and floated at the surface of each mesocosm.



were observed actively swimming in the water column throughout the experiment and appeared active in the benthos during the diver survey. Although less abundant, *Cerianthopsis* were observed with their tentacles extended and considered active in all systems. Benthic organisms are not prey items for larval flounder and the abundance of benthic predators was the same prior to the start and at the end of the study. In addition to the benthic predators, chaetognaths and *Sarsia tubulosa* medusae, known or suspected pelagic predators on larval fish (Khulmann 1977; Northeast Utilities 1999), were also present in some mesocosms (Table 4). No pelagic predators were collected in plankton or pumped samples throughout the experiment but were present in the ring and overflow nets when the tanks were drained at the end of the study. The relationship between daily instantaneous larval mortality rates and the abundance of predators (benthic plus pelagic) is highly significant, with high mortality rates correlated with abundant active predators (Fig. 10).

Discussion

Winter flounder has historically been a dominant demersal fish species collected in Narragansett Bay (Jeffries and Terceiro 1985). Commercial catch and recreational catch have declined severely since 1979 (Gibson 1998). In 1990, our Narragansett Bay ichthyoplankton survey indicated that winter flounder larvae were significantly less abundant in all regions of the Bay compared with an earlier survey in 1972–1973 (Keller et al. 1999b). Because of concerns over drastic decreases in abundance, the Rhode Island Department of Environmental Management banned fishing for winter flounder in Narragansett Bay and its adjacent salt ponds in 1991. A limited fishery was reopened in 1995, with very little improvement in catch. Although winter flounder have tended to be somewhat cyclical throughout historical assessments, each peak and trough is now lower than preceding values and troughs are extending over longer time periods (Gibson 1998).

Although overfishing plays a role in the population decline, other factors are important in controlling recruitment

Fig. 8. Mean (\pm range) winter flounder larval standard length for each sample period over the experimental period in the individual mesocosm. (A–C) Warm tanks; (D–F) cool tanks.

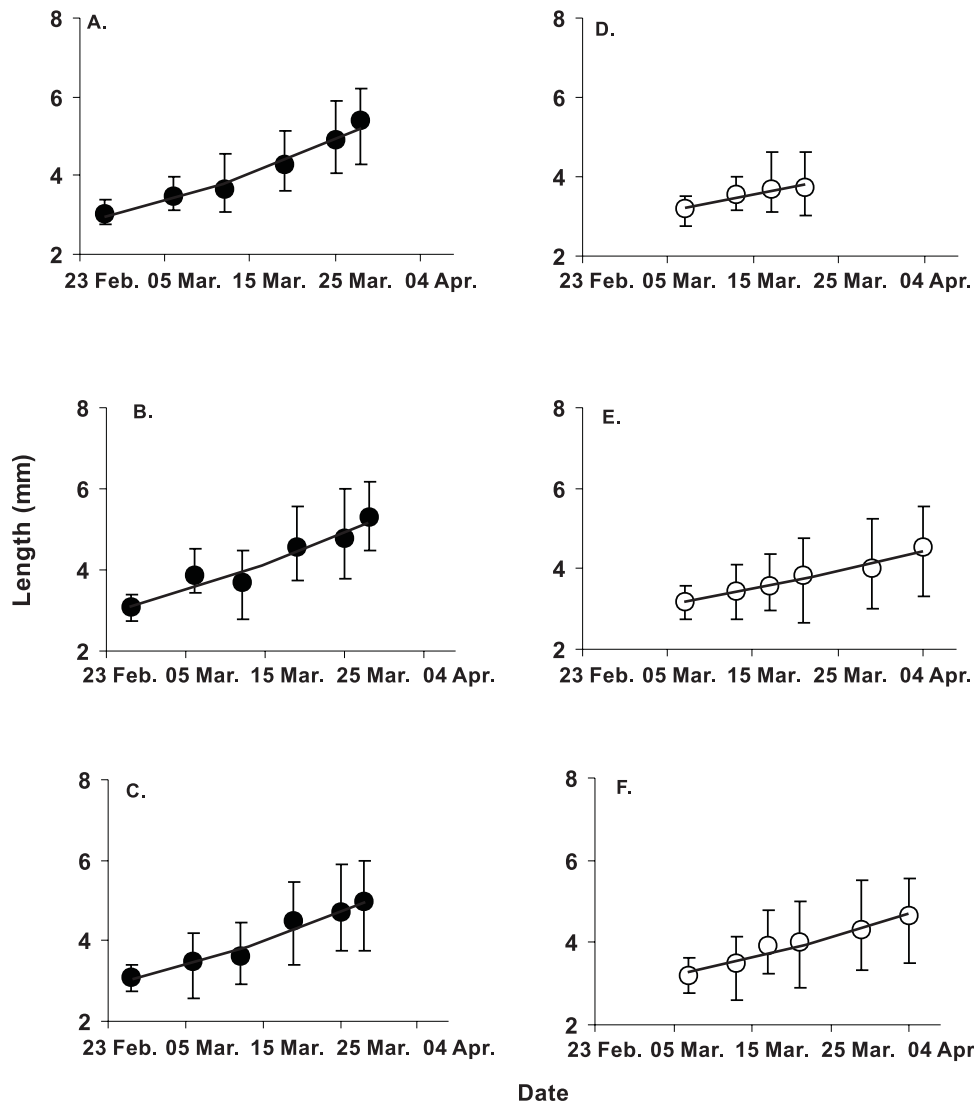


Table 2. Average daily instantaneous growth rates (G) and associated statistics for winter flounder larvae in each mesocosm from eq. 1.

Treatment	G (\pm SE) (day^{-1})	n	r^2	P	Δt (days)
Warm, 3	0.018 (0.001)	125	0.88	0.0001	31
Warm, 7	0.018 (0.001)	80	0.92	0.0001	31
Warm, 9	0.016 (0.001)	161	0.75	0.0001	31
Cool, 5 ^a	0.011 (0.001)	49	0.61	0.0001	16
Cool, 9	0.013 (0.001)	105	0.85	0.0001	26
Cool, 13	0.012 (0.001)	146	0.78	0.0001	26

Note: Mesocosm number is shown next to the treatment. Values in parentheses are standard errors for the estimated parameters. Treatments are defined in the text.

^aData collection ceased in this mesocosm following the spill of egg basket on 21 March.

of winter flounder. Annual abundance, when appropriately lagged (most flounder take 1–2 years to enter the catch), is negatively correlated with winter temperature, indicating

that warmer temperatures during spawning result in fewer fish (Jeffries and Johnson 1974; Jeffries and Terceiro 1985; Northeast Utilities 1999). Buckley et al. (1990) noted that cold winters favored good survival by facilitating production of large larvae in good condition (high RNA and protein content). Jeffries et al. (1989) presented the hypothesis that temperature controls flounder abundance through predation. Earlier mesocosm studies designed to assess the importance of eutrophication on growth and survival of winter flounder larvae instead suggested that benthic predators or competitors play an important role in mortality of winter flounder larvae (Klein-MacPhee et al. 1993). Since winter water temperatures in Narragansett Bay have increased significantly over the past 40 years, understanding the impact of elevated temperature on winter flounder recruitment is crucial. The current experiment was designed to test the hypothesis that experimentally increasing temperature would produce a detectable change in the survival of winter flounder larvae to metamorphosis. We hypothesized that the observed decline in winter flounder abundance was a result of the interaction

Fig. 9. Mean abundance (± 1 SE) of winter flounder larvae in (A) the warm treatments and (B) the cool systems over the experimental period.

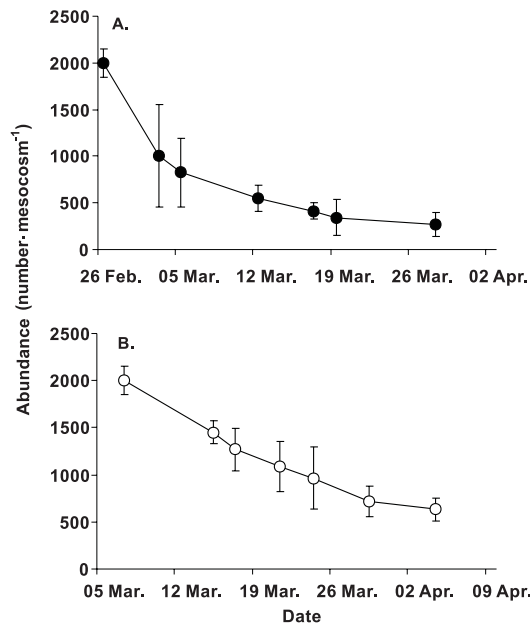


Table 3. Average daily instantaneous mortality rates (Z) and associated statistics for winter flounder larvae in each mesocosm from eq. 2.

Treatment	Z (\pm SE) (day^{-1})	n	r^2	P
Warm, 3	0.058 (0.019)	7	0.60	0.04
Warm, 7	0.093 (0.031)	7	0.63	0.03
Warm, 13	0.068 (0.008)	7	0.93	0.0004
Cool, 5 ^a	0.037 (0.019)	5	0.93	0.007
Cool, 9	0.040 (0.008)	7	0.81	0.006
Cool, 15	0.041 (0.006)	7	0.89	0.0004

Note: Mesocosm number is shown next to the treatment. Values in parentheses are standard errors for the estimated parameters. Treatments are defined in the text.

^aData collection ceased in this mesocosm following spill of egg basket on 21 March.

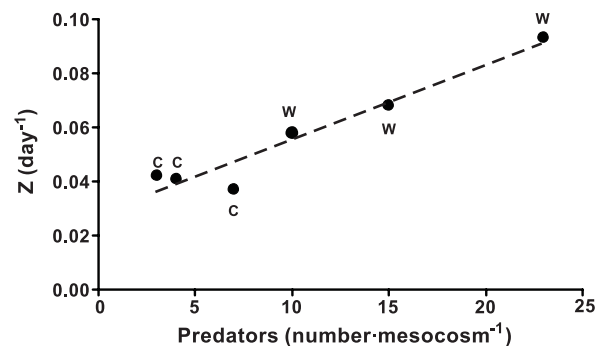
between temperature and either food availability or predation, the primary factors affecting larval fish recruitment (Cowan and Houde 1990). The above results indicated that temperature differences at the experimental level impacted winter flounder larval growth and survival. Our results demonstrated that warmer winter water temperature influences the food web leading to winter flounder as well as egg mortality, percent hatch, size at hatch, growth rate, and larval mortality. It is important to note when interpreting the experimental results that although water temperature was increased 3.25°C relative to controls, the actual increase relative to the Bay temperature over the same period was 0.7°C. In essence, the experiment is comparing current and future increases in winter temperatures resulting from global climate change with temperatures more typical of cooler winters in the past.

Table 4. Abundance (number·mesocosm⁻¹) of benthic and pelagic predators in each mesocosm throughout the experiment.

Treatment	<i>Crangon</i> sp.	<i>Cerianthopsis</i> sp.	Chaetognaths	<i>Sarsia</i> sp.
Warm, 3	4	5	0	1
Warm, 7	18	2	1	2
Warm, 13	5	9	1	0
Cool, 5	3	5	0	2
Cool, 9	0	4	0	0
Cool, 15	2	3	0	0

Note: Mesocosm number is shown next to the treatment. Although present in the cool systems, *C. septemspinosa* were inactive and not included in the total predator abundance utilized in the analysis of mortality rates.

Fig. 10. Empirical relationship ($Y = 0.003X + 0.028$, $r^2 = 0.93$) between larval winter flounder daily instantaneous mortality rate (Z) and the abundance of active predators in the warm (W) and cool (C) treatments.



Long-term data indicate a significant increase in water temperature in Narragansett Bay during February–April over the past 40 years. Temperatures in the warm mesocosms were similar to those observed in Narragansett Bay during much of the experiment and typical of late winter temperatures generally observed in recent years. Temperatures in the cool systems were closer to those observed during recent cold winters (e.g., 1996) and more typically in the past. Both warm and cool systems demonstrated the gradual temperature increase over time that occurs from February through early April and were within the range of temperatures (–1.8 to 15°C) at which winter flounder larvae are known to occur (Williams 1975; Rogers 1976).

In an earlier temperature experiment that did not include ichthyoplankton (Keller et al. 1999a), we observed that the characteristic winter–spring bloom failed to occur in warm systems, similar to its failure to appear in the Bay in recent years. In the present study, when the mesocosms were filled during a period with moderate Chl *a* levels in the Bay (~15 $\mu\text{g}\cdot\text{L}^{-1}$), blooms developed in both warm and cool systems. These results suggest that the moderate Chl *a* levels present in the tanks at the time of the fill were sufficient to prevent grazers from initially controlling bloom development. Towards the end of the study, we observed an increase in phytoplankton in the cool systems similar to our earlier results (Keller et al. 1999a). The dominant phytoplankton species present during the experiment (*Thalassiosira* spp., *Chaetoceros* spp., and *S. costatum*) are those typically

observed during the winter–spring bloom period in the parent system, Narragansett Bay (Smayda 1973). The phytoplankton abundance and biomass levels are typical of the levels occurring in the upper regions of Narragansett Bay where winter flounder larvae are most abundant (Durbin and Durbin 1981; Keller et al. 1999b).

Copepod abundance increased in the warm systems compared with cool systems, again similar to results observed in our prior temperature experiment when flounder larvae were not present (Keller et al. 1999a). The rapid increase in copepod nauplii in the warm systems reflects the temperature-mediated hatching rate of *A. hudsonica* resting eggs combined with an egg production rate of up to 42 eggs·female⁻¹·day⁻¹ (Sullivan and McManus 1986). The dominant copepod species present were those typically observed in Narragansett Bay in February and March and at levels comparable with those occurring in the upper Bay (Durbin and Durbin 1981). Winter flounder larvae feed primarily on diatoms, dinoflagellates, copepod nauplii, and copepodites (calanoid and harpacticoid) (Laurence 1977; Klein-MacPhee et al. 1993). The net result of the changes in phytoplankton and copepod abundance in the warm systems relative to the cool was that more food was available for winter flounder larvae in the warm systems. Our highest survivals during the present study occurred in the systems with the lowest food supply. There was no relationship between survival and food abundance in the current study or in our studies conducted during 1988–1991 (Klein-MacPhee et al. 1993). Laurence (1977) found that winter flounder larvae failed to survive beyond 2 weeks at prey densities between 10 and 100 nauplii·L⁻¹ in laboratory studies. During the present study, copepod nauplii abundance in cool systems ranged from 2 to 27·L⁻¹, while in warm systems, nauplii were more abundant, 3–35·L⁻¹. Despite the fact that these values fall within the range associated with 100% mortality at 2 weeks in Laurence's (1977) study, we observed relatively high survival in all systems. Research in other mesocosms has also demonstrated that fish larvae grow and survive with food levels lower than laboratory estimates of required amounts (Øiestad 1985; Cowan and Houde 1990). Although food availability was not a primary factor affecting mortality in the current study, the potential for variability in food quality between warm and cool systems does exist. Recall that *Skeletonema* and *Chaetoceros* spp. were more abundant in cool systems, while *Thalassiosira* spp. were more abundant in warm systems. The dominant copepod was the same species (*A. hudsonica*) in both treatments. The nutritional value of *Skeletonema* and *Chaetoceros* versus *Thalassiosira* is unknown. Although temperature had a significant impact on the food web leading to winter flounder, this does not appear to be the dominant factor influencing larval mortality during the study.

Temperature had a direct impact on the incubation period in the current study, with cooler temperature resulting in a longer incubation. Previous research on winter flounder from Narragansett Bay has also indicated that incubation time of eggs varies inversely with temperature (Rogers 1976). Rogers (1976) noted that time to hatching was approximately 4 weeks at 3°C and 2 weeks at 8°C. Our values for the warm systems fall between these, with hatching occurring at about 3 weeks at 4.1°C. In the cool systems with

average incubation temperatures of 1.6°C, the incubation period was slightly longer than the 4-week period described by Rogers (1976) at 3°C.

Winter flounder larvae hatched at a larger size in the cool mesocosms relative to the warm. Buckley et al. (1990) similarly noted that larvae were larger when incubated at 2°C compared with larvae incubated at warmer temperatures (7°C). Although smaller at hatching, the larvae in the warm mesocosms grew more rapidly. The difference in growth rates between treatments may be interpreted as a density-dependent effect, since the higher mortality in warm systems resulted in a lower abundance of fish there. Daily instantaneous growth and mortality rates were significantly and inversely related ($r^2 = 0.70$). The daily instantaneous growth rates observed in the warm systems (0.016–0.018·day⁻¹) were not significantly different from the rates observed in an earlier unrelated study (0.019–0.022·day⁻¹) at somewhat warmer temperatures (7.8°C) (Klein-MacPhee et al. 1993), while those from the cool systems (0.011–0.013·day⁻¹) were significantly lower.

Capture efficiency was unrelated to treatment and within the range expected for larval winter flounder (Pearcy 1962) and observed in prior mesocosm studies (G. Klein-MacPhee, unpublished data). Mortality rates in cool systems (0.037–0.041·day⁻¹) fell within the range of values previously observed in mesocosms operated without a benthos (0.002–0.045·day⁻¹) (Klein-MacPhee et al. 1993). These values approximate rates more common for laboratory studies where there is no predation. The higher and more variable rates observed in the warm systems (0.058–0.093·day⁻¹) tended to be somewhat lower than rates previously observed in our experiments with a benthos present (0.100–0.193·day⁻¹). Even in warm systems, the abundance of predators was relatively low during the temperature experiment. The maximum density of *Crangon* in the experiment was ~5·m⁻² and well below the maximum winter density observed by Thornton-Whitehouse (1994) in Narragansett Bay (80·m⁻²). *Cerianthopsis* density was also low in the mesocosms relative to densities observed in the field (Holohan 1993). The variability in mortality rates was linearly correlated with the abundance of predators. A linear relationship between mortality rate and abundance of predators was unexpected and may be a result of the low numbers of predators present.

The cumulative impact of warmer temperatures on the early life history of winter flounder from hatching through the late larval stage resulted in 10–16% fewer larvae surviving to metamorphosis (~6 weeks). These results together with the potential for increased egg predation and post-settlement predation, most notably by *Crangon* (van der Veer and Bergman 1987; Whitting 1993), as a result of elevated winter water temperature suggest that temperature and its impact on predation rate may play a major role in regulating winter flounder abundance. Now that we know that this temperature-mediated predation effect exists, regional managers should attempt to incorporate the potential impact of warm winters in their management plans. One such approach would be to reduce fishing pressure following periods of successive warm winters. Currently, chronic overexploitation is associated with a long-term decline in winter flounder stock abundance despite periodic production of good

year-classes (Gibson 1998), suggesting that even with the higher recruitment associated with cooler winters, overfishing is a problem in Narragansett Bay.

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References

- Buckley, L.J., Smigielski, A.S., Halavik, T.A., and Laurence, G.C. 1990. Effects of water temperature on size and biochemical composition of winter flounder *Pseudopleuronectes americanus* at hatching and feeding initiation. *Fish. Bull. U.S.* **88**: 419–428.
- Cowan, J.H., and Houde, E.D. 1990. Growth and survival of bay anchovy *Anchoa mitchilli* larvae in mesocosm enclosures. *Mar. Ecol. Prog. Ser.* **68**: 47–57.
- Crawford, R. 1990. Winter flounder in Rhode Island coastal ponds. National Seagrant Depository Publication RIU-G-90-001, Narragansett, R.I.
- Durbin, A.G., and Durbin, E.G. 1981. Standing stock and estimated production rates of phytoplankton and zooplankton in Narragansett Bay, Rhode Island. *Estuaries*, **4**: 24–41.
- Gibson, M.R. 1998. Recent trends in abundance, recruitment, and fishing mortality for winter flounder in Narragansett Bay and Rhode Island coastal waters. Report to the Rhode Island Marine Fisheries Council. Res. Ref. Doc. 98, Rhode Island Division of Fish and Wildlife, Wickford, R.I.
- Holohan, B.A. 1993. Population density and grazing of *Cerianthiopsis americanus* in Narragansett Bay. M.S. thesis, University of Rhode Island, Kingston, R.I.
- Houde, E.D. 1987. Fish early life dynamics and recruitment variability. *Am. Fish. Soc. Symp.* **2**: 17–29.
- Jeffries, H.P., and Johnson, W.C. 1974. Seasonal distribution of bottom fishes in the Narragansett Bay area: seven-year variations in the abundance of winter flounder (*Pseudopleuronectes americanus*). *J. Fish. Res. Board Can.* **31**: 1057–1066.
- Jeffries, H.P., and Terceiro, M. 1985. Cycle of changing abundances in the fishes of the Narragansett Bay area. *Mar. Ecol. Prog. Ser.* **25**: 239–244.
- Jeffries, H.P., Keller, A.A., and Hale, S. 1989. Predicting winter flounder (*Pseudopleuronectes americanus*) catches by time series analysis. *Can. J. Fish. Aquat. Sci.* **46**: 650–659.
- Keller, A.A., Oviatt, C.A., Walker, H.A., and Hawk, J.D. 1999a. Predicted impact of elevated temperature on the magnitude of the winter–spring phytoplankton bloom in temperate coastal waters: a mesocosm study. *Limnol. Oceanogr.* **44**: 344–356.
- Keller, A.A., Klein-MacPhee, G., and St. Onge-Burns, J. 1999b. Abundance and distribution of ichthyoplankton in Narragansett Bay, Rhode Island, 1989–1990. *Estuaries*, **22**: 149–163.
- Khulmann, D. 1977. Laboratory studies on the feeding behavior of the chaetognaths *Sagitta setosa* J. Muller and *S. elegans* Verrill with special reference to fish eggs and larvae as food organisms. *Ber. Dtsch. Wiss. Komm. Meeresforsch.* **25**: 163–171.
- Klein-MacPhee, G., Howell, W.H., and Beck, A.D. 1982. Comparison of a reference strain and four geographic strains of *Artemia* as food for winter flounder, *Pseudopleuronectes americanus*, larvae. *Aquaculture*, **29**: 279–284.
- Klein-MacPhee, G., Sullivan, B.K., and Keller, A.A. 1993. Using mesocosms to assess the influence of food resources and toxic material on larval fish growth and survival. *Am. Fish. Soc. Symp.* **14**: 105–116.
- Laurence, G.C. 1974. Growth and survival of haddock (*Melanogrammus aeglefinus*) larvae in relation to planktonic prey concentration. *J. Fish. Res. Board Can.* **31**: 1415–1419.
- Laurence, G.C. 1977. A bioenergetic model for the analysis of feeding and survival potential of winter flounder, *Pseudopleuronectes americanus*, larvae during the period from hatching to metamorphosis. *Fish. Bull. U.S.* **75**: 529–546.
- Laurence, G.C., Halavik, T.A., Burns, B., and Smigielski, A.S. 1979. An environmental chamber for monitoring ‘in situ’ growth and survival of larval fishes. *Trans. Am. Fish. Soc.* **108**: 197–203.
- Lorenzen, C.J. 1966. A method for continuous measurement of *in vivo* chlorophyll concentration. *Deep-Sea Res.* **13**: 223–227.
- Northeast Utilities. 1999. Monitoring the marine environment of Long Island Sound at the Millstone Nuclear Power Station, Waterford, CT. Northeast Utilities, Environmental Laboratory, Waterford, Conn.
- Øiestad, V. 1985. Predation on fish larvae as a regulatory force, illustrated in mesocosm studies with large groups of larvae. *NAFO Sci. Counc. Stud.* **8**: 25–32.
- Oviatt, C.A. 1994. Biological considerations in marine enclosure experiments: challenges and revelations. *Oceanography*, **7**: 45–51.
- Oviatt, C.A., Keller, A.A., Sampou, P.A., and Beatty, L.L. 1986. Patterns of productivity during eutrophication: a mesocosm experiment. *Mar. Ecol. Prog. Ser.* **28**: 69–80.
- Pearcy, W.G. 1962. Ecology of young winter flounder in an estuary. *Bull. Bingham Oceanogr. Collect. Yale Univ.* **18**: 1–78.
- Rogers, C.A. 1976. Effects of temperature and salinity on the survival of winter flounder embryos. *Fish. Bull. U.S.* **74**: 52–58.
- Schuermans, C.J.E. 1995. The world heat budget: expected changes. *In* *Climate change: impact on coastal habitation*. Edited by D. Eisma. Lewis Publishers, CRC Press, Boca Raton, Fla. pp. 1–16.
- Smayda, T.J. 1973. The growth of *Skeletonema costatum* during a winter–spring bloom in Narragansett Bay, Rhode Island. *Norw. J. Bot.* **20**: 219–247.
- Sournia, A. (Editor). 1978. *Phytoplankton manual*. UNESCO, Paris.
- Sullivan, B.K., and McManus, L.T. 1986. Factors controlling seasonal succession of the copepods *Acartia hudsonica* and *A. tonsa* in Narragansett Bay, Rhode Island: temperature and resting egg production. *Mar. Ecol. Prog. Ser.* **28**: 121–128.
- Thornton-Whitehouse, S. 1994. The abundance and distribution of *Crangon septemspinosa* in Narragansett Bay. Ph.D. thesis, University of Rhode Island, Kingston, R.I.
- van der Veer, H.W., and Bergman, M.J.N. 1987. Predation by crustaceans on a newly settled 0-group plaice *Pleuronectes platessa* population in the western Wadden Sea. *Mar. Ecol. Prog. Ser.* **35**: 203–215.
- Whitting, D.A. 1993. Effects of body size on probability of predation for juvenile summer and winter flounder based on laboratory experiments. *Fish. Bull. U.S.* **91**: 577–581.
- Williams, G.C. 1975. Viable embryogenesis of the winter flounder *Pseudopleuronectes americanus* from –1.8° to 15°C. *Mar. Biol.* **33**: 71–74.
- Winer, B.J. 1971. *Statistical principles in experimental design*. 2nd ed. McGraw-Hill, New York.