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Detecting critical periods in larval flatfish populations

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Abstract

We evaluate the time-course of deaths and evidence of periods of increased mortality (i.e., critical periods) in laboratory populations of larval flatfish. First, we make the distinction between age-at-death and abundance-at-time data for fish larvae, the latter being typical in studies of natural populations. Next, we describe an experimental investigation of age- and temperature-dependent mortality in larval winter flounder, *Pseudopleuronectes americanus*. The survivorship curves of these populations differed significantly in both the magnitude and time-course of mortality among the four water temperatures evaluated (7, 10, 13, and 16℃). Mortality was highest in the cooler temperatures and concentrated in the third quarter of larval life, largely concurrent with settlement of surviving members of the cohort. Among the statistical methods for analysing survival data, the proportional-hazards model with time-varying covariates proved best at capturing the patterns of age-specific mortalities. We conclude that fair appraisals of recruitment hypotheses which are predicated on periods of high, age-specific mortality that vary with environmental conditions (e.g., Hjort's critical period hypothesis) will require: (1) data that are based on age, not time; (2) data that are of higher temporal resolution than commonly available at present and (3) analytical methods that are sensitive to irregularities in survivorship curves. We suggest four research approaches for evaluating critical periods in nature. © 2001 Elsevier Science B.V. All rights reserved.

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1. Introduction

Fisheries biologists have long sought to understand fluctuations in fish populations by focusing on the factors responsible for the high and variable levels of mortality that occur during the early life-stages of fish (Gulland, 1965). One potential source of mortality for young larvae is the failure to establish feeding. Low prey densities or reduced capabilities in capturing prey could result in starvation of larvae or in weakened individuals that are more susceptible to predation and other sources of mortality (Anderson,

1988). Many studies purport to evaluate whether mortality is indeed concentrated at ages when young larvae initiate feeding and, by implication, is due to starvation as suggested by Hjort's critical period hypothesis (Hjort, 1914). Reviews of those studies by Marr (1956), May (1974), and Leggett and DeBlois (1994) conclude that data on larval fish abundances either do not support a link between starvation mortality near the time of first feeding and the level of recruitment or are inconclusive with respect to this hypothesis.

We argue that many prior studies share a fundamental limitation in their attempts to determine whether high levels of mortality punctuate the ages at first feeding or other age intervals in larval life.

These intervals of relatively high mortality are here-after referred to as 'critical periods' (Vladimirov, 1975; Browman, 1989; Thorisson, 1994). Ages at death are the preferred demographic data with which to examine the time-course of mortality and its associ-ation with life history features (Keyfitz, 1968). Such data are routinely estimated from abundances of annual age-classes for populations of adult fish. For larval populations, especially with respect to detecting critical periods, the ideal data would be mortalities (or abundances) at daily or near-daily age resolutions. If available, such data could be used to construct survivorship curves that reflect the true, age-specific mortality schedules for natural populations. Dead larvae, however, are rarely encountered in nature (Strasburg, 1958; Wiborg, 1976). Due to the recent-ness and requirements of larval aging techniques, dead larvae have not typically been aged. Ichthyo-plankton studies usually sample larvae of mixed ages and report estimates of larval abundances-at-time (Smith and Richardson, 1977). Undoubtedly, abundance-at-time data reflect the production and loss of individuals in a population to a degree, but these data are not equivalent to abundance-at-age and age-at-death (or life span) data and do not share the same utility for detecting critical periods. These distinctions are fundamental to correctly assessing critical periods and will be demonstrated in this paper.

Abundance-at-time data for larval fish, as generated in catch curves for a cohort, often show a negative exponential decline. This tendency towards a simple decay pattern is even more likely when abundance data are pooled across life stages, locations, or years in an attempt to broaden results on patterns of mor-tality (see examples in Dahlberg, 1979). It has long been appreciated that simple functions applied to abundance-at-time data of coarse temporal resolution overlook important interim events (Ricker, 1975). The apparent suitability of a one-parameter decay curve, and indeed the magnitude of the estimated instant-aneous mortality, is known to vary with the frequency at which populations are sampled (Taggart and Frank, 1990). Therefore, we view a successful fit of an expo-nential decay model to abundance-at-time data either for sub-stages or for the entirety of larval life as being of limited value in identifying age-specific critical periods.

1.1. Abundance-at-age versus abundance-at-time: a hypothetical example

We present a simple example based on a set of hypothetical life spans (Fig. 1) in order to demonstrate how rapidly the details of a true, age-based survivor-ship curve are lost when a mixed-age or composite population is sampled. The survivorship curve corre-sponding to these life spans (i.e., the termini of ranked life spans, Fig. 1) possesses the following details. After initial high survival (Fig. 2(a), 'I'), a period of precipitous mortality occurs ('II') followed by a period of low and constant mortality ('III'). Mortality is once again relatively high for a brief period ('IV') and lessens thereafter ('V'). Two-thirds of the popu-lation is lost in each period of high mortality, meaning that the mortality risk is the same for fish that began each of the two critical periods.

Next, we create a mixed-age population of larvae, as would be typical of most natural populations wherein larvae hatch over an interval of days to weeks. Our mixed-age population consists of 20 consecutive daily cohorts, each beginning with
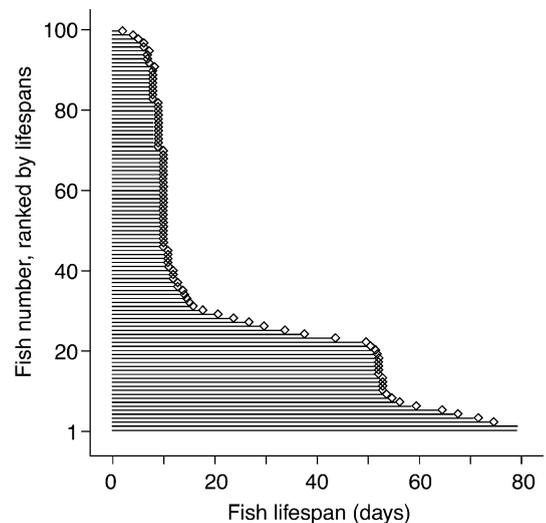

Fig. 1. Hypothetical data on life span, age-specific mortality, and survivorship of a cohort of fish larvae. Life spans (horizontal lines) and ages at death ($\diamond$) are shown for a cohort of 100 larvae that hatched on the same day. Life spans are placed in rank order (long-est life span first, shortest life span last). The pattern of ranked ages at death is the cohort survivorship curve, expressed as the number of individuals alive at age.
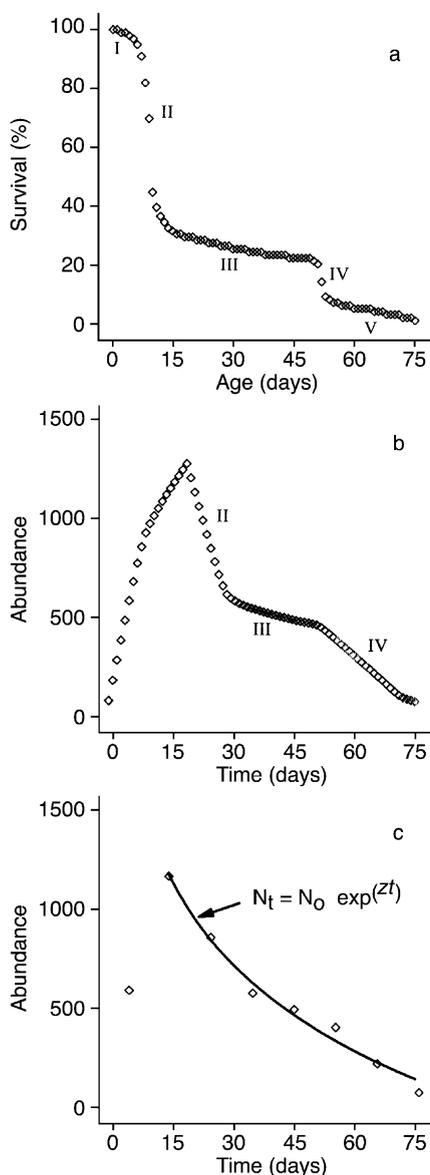
Fig. 2. Hypothetical data on survivorship, abundance, and sampled abundance of fish larvae. (a) Survivorship curve with periods of low (I, III, V) and high (II, IV) mortality for a daily cohort that began with 100 fish larvae (from Fig. 1, expressed as percentage alive at age). (b) Abundance over time of a population comprised of 20 consecutive daily cohorts, each with initial abundances and survivorship curves identical to that in a. (c) Sampled abundances of population in b, sampled every 10 days beginning on day 5. An exponential decay curve is fitted to the region of decreasing abundances.

100 individuals and each experiencing identical age-specific mortality schedules (Fig. 2(a)). The underlying survivorship curves are poorly reflected in the resulting plot of abundance-at-time (Fig. 2(b)). The intensity and duration of the two critical periods (Fig. 2(b), 'II', 'IV') are masked and the long period of low mortality ('III') becomes a shortened period of moderate decline in abundances.

Lastly, in order to place our hypothetical example in a more realistic context, we depict estimated abundance-at-time data as would be gathered from sampling the population. To do so, we sampled the hypothetical population every 10 days starting at day 5. The resulting sampled data only weakly resemble the underlying cohort survivorship curves; indeed, an exponential decay model fits the declining portion of the production curve well and would support the conclusion that no evidence exists for one, let alone two critical periods (Fig. 2(c)).

This demonstration used hypothetical mortality data that are exceptionally simple compared to those that would be collected for natural populations. In our example, the input of hatching larvae was continuous and uniform for 20 days, the age-specific mortality was repeated exactly for all daily cohorts, and the population was sampled without error or bias. Even under these ideal conditions, we could not argue for the existence of a critical period. Based on this example, we believe that it would be exceedingly unlikely to detect an age-specific critical period in natural populations comprised of mixed-age larvae if one relies solely on abundance-at-time data.

1.2. Prior experimental approaches to studies of larval mortality

An alternative to using field-based abundance-at-age data to evaluate the occurrence of critical periods is to monitor mortality in laboratory populations. Although such studies cannot directly assess links to recruitment, they do offer a potential means of illuminating the pattern and mechanisms of processes that may result in recruitment variability in nature.

Many laboratory-based rearing studies that report larval survival are conducted in one of three ways. First, survival is part of a suite of characteristics measured for the population being reared. It is estimated at an aggregate level and is based on

the proportion of the initial number of larvae that survives to the termination of the study (Houde, 1977; Chambers and Leggett, 1987; Buckley et al., 1991). This 'numbers in–numbers out' approach does not address the time-course of mortality.

Second, survival is a primary response monitored in short-term experiments directed towards assessing the effects of food deprivation after hatching on larval growth, condition, and survival. Examples include 'point-of-no-return' studies (Blaxter and Hempel, 1963; Gadomski and Petersen, 1988; Bisbal and Bengtson, 1995), and studies of 'irreversible starvation' (Lasker et al., 1970; May, 1971; McGurk, 1984). These studies monitor populations at high frequencies for mortalities, and thus the time-course of larval mortality is revealed, but only until shortly after first feeding.

A third approach records age-specific mortalities for the entirety of larval life. This requires the investigator to be able to see, count, and remove mortalities on a regular basis. The resulting data on the time-course of mortality can be viewed as a representation of an intrinsic population survivorship curve under the prevailing environmental conditions. Prior studies of this type have often been descriptive, small-scale, pilot studies directed towards aquaculture (Shelbourne, 1975; Jones et al., 1981). Of the three approaches, we contend that this one is appropriate for detecting critical periods. It is the basis of the experiments reported here.

Our objectives in the present investigation were to: (1) experimentally generate high-resolution age-at-death data for larval flatfish from different environmental conditions, (2) identify the appropriate statistical models with which to describe and analyze these data and (3) interpret the patterns of age-specific mortalities in light of hypothesized critical periods for larval fish. We assess the magnitude and time-course of mortality during larval life of winter flounder, *Pseudopleuronectes americanus*, and examine how age-specific patterns of larval mortality are affected by water temperature. We also suggest several research approaches for future analysis of critical periods of larval fish populations in nature.

2. Methods

The larval winter flounder used as experimental subjects were the products of crosses between two females and one male winter flounder that were collected in Sandy Hook Bay, New Jersey, USA. One hundred, 1-day-old winter flounder larvae were allocated to each of eight, 8-dm^3 rearing containers. These containers had previously been filled with 6 dm^3 of filtered and sterilised seawater that was conditioned with unicellular marine algae (*Isochrysis galbana*) prior to the allocation of the larvae. Two containers were distributed to each of four experimental baths that were maintained at 7, 10, 13, or 16°C, which represent temperatures that winter flounder larvae might encounter in nature. The resulting experiment was a twice replicated, single-factor design with different paternal half-sibships constituting the replicates. Prey were added to containers daily; rotifers (*Brachionus plicatilis*) at 5 per cm^3 throughout larval life and brine shrimp (*Artemia* sp.) at 1 per cm^3 after first evidence of larvae in each container beginning to settle on the container bottom.

Containers were inspected daily for mortalities and evidence of first settlement. Once individuals were observed lying on the container bottom, containers were also examined daily for metamorphosed fish. In order to reduce error in determining the terminus of larval life, we defined metamorphosis as attainment of developmental stage H, which is signified in these flatfish as the time at which the migrating eye reaches the crest of the dorsum (see Minami, 1982; Seikai et al., 1986 for staging characteristics). Individuals approaching stage H were removed from their rearing containers, inspected microscopically at 6× to assess developmental stage, and either placed in a new container for juveniles or returned to their original larval container if they had not yet attained stage H. Individuals that had metamorphosed had also, by definition, survived larval life. These survivors were registered as right-censored observations in the survival analyses described below. Deaths that occurred within 48 h of the initiation of the experiment (range, 8–22 individuals per population) were considered as caused by experimental handling, not starvation, because larvae of this age have not yet depleted their yolk reserves (Chambers et al., unpublished data). Handling mortalities were excluded in subsequent survival analyses.

Various types of models are available to describe and compare populations with respect to the time-course of events (here, mortalities). We evaluated
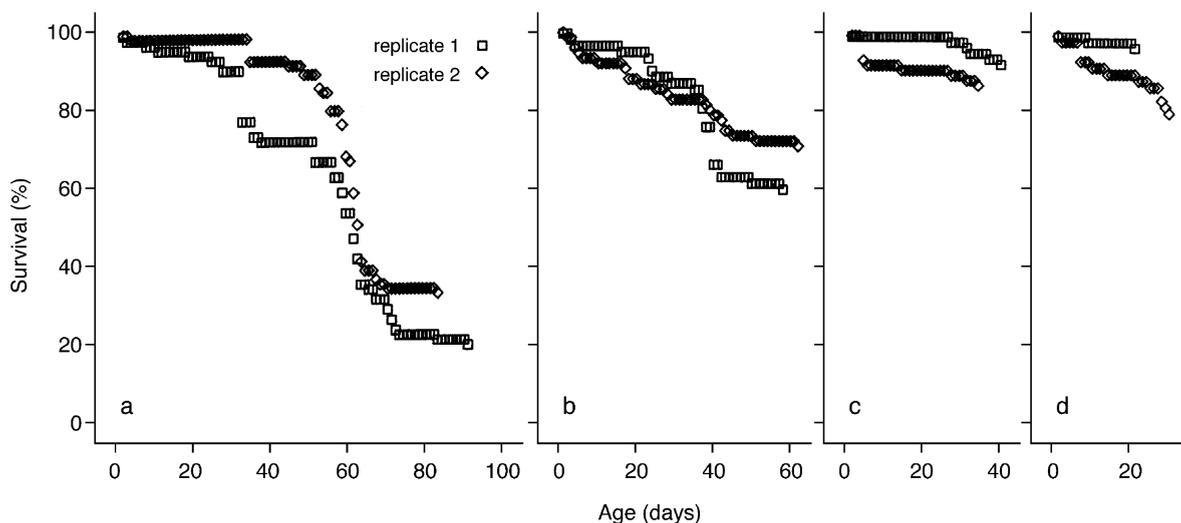
Fig. 3. Survivorship curves for replicate populations of larval winter flounder, *Pseudopleuronectes americanus*, reared in the laboratory from hatching through metamorphosis (stage H) at constant temperatures of (a) 7°, (b) 10°, (c) 13°, and (d) 16°C. At the terminus of each survivorship curve all larvae had either died or metamorphosed.

the following four models for their adequacy in depicting our experimentally generated survivorship curves: (1) exponential decay, (2) accelerated failure time, (3) proportional hazard and (4) proportional hazard with time-dependent covariates. The first two models are parametric in form and the last two are partly parametric; they permit covariates such as a temperature treatment but do not subsume an underlying error structure. The exponential-decay model (i.e., the $N_t = N_0 e^{zt}$ formulation) is used here primarily as a reference point. It is commonly used in fisheries research to represent declines in fish abundances during the early life history in total or during each of several sequential stages within larval life. The accelerated-failure-time model has two parameters that reflect the location and shape of the statistical distribution of survival data. We assumed a Weibull error structure for this model. The two proportional-hazards models allow for covariates, in this case water temperature, and the time-dependent version permits the populations being compared to differ in the time-course of their mortalities. Beyond providing estimates of survivorship curves, these models also generate probability-density functions of mortality and age-specific mortality risks for the population. Details on these models are in Kalbfleisch and Prentice (1980), Chambers and Leggett (1989), and Allison (1995).

We pooled observations among replicates within temperatures prior to our evaluation of the alternative survival models. Pooling was supported by the lack of significant replicate-within-temperature effects on the survivorship curves (Wald's $X^2 = 1.04$, $df = 1$, $p > 0.31$). Analyses were conducted with SAS and SYSTAT survival procedures (Allison, 1995; SYSTAT, 1999).

3. Results

Survival through larval life ranged from 20 to 97% of the initial number of larvae established in a population. The two replicate populations within a rearing temperature were consistent with one another with respect to their survivorship curves, but the magnitude and time-course of mortality varied among the experimental temperatures (Fig. 3). There was no obvious evidence of a critical period corresponding to the time of yolk-sac depletion and first feeding. Yolk depletion and first feeding of larvae occurred at approximately 3–10 days post-hatch, varying inversely with temperature (Chambers et al., unpublished data). Populations at the coolest temperature (7°C) exhibited the greatest mortality (Fig. 3(a)). Most of the mortality in the 7°C populations occurred in the
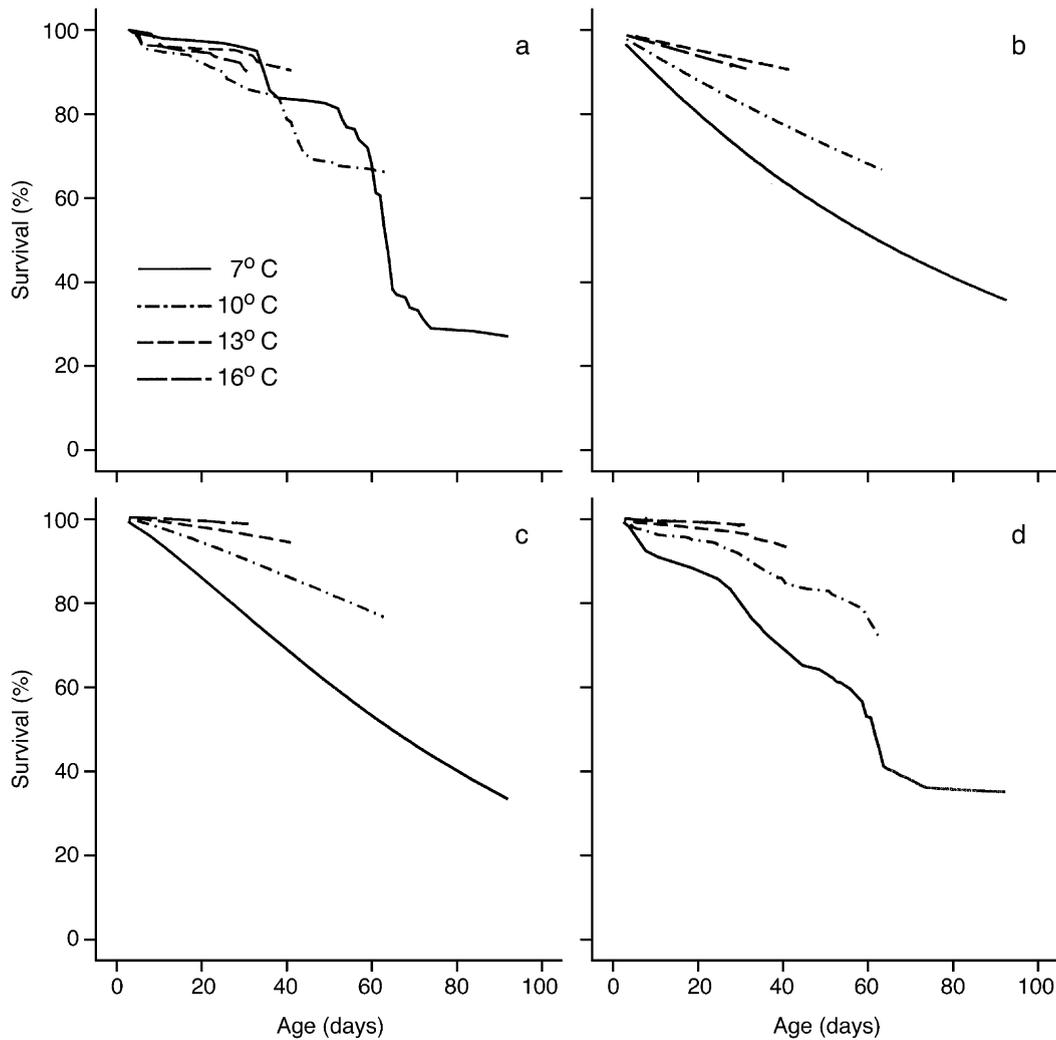
Fig. 4. Actual and predicted survivorship curves for laboratory populations of larval winter flounder, *Pseudopleuronectes americanus*, reared in the laboratory from hatching through metamorphosis (stage H) at constant temperatures of 7, 10, 13, and 16°C. (a) Actual survivorship curves for all temperatures (replicates pooled). (b) Predicted survivorship curves from an exponential-decay model. (c) Predicted survivorship curves from an accelerated-failure-time model with Weibull error structure. (d) Predicted survivorship curves from a proportional-hazards model with temperature as a covariate.

third quarter of larval life (days 55–70) although a modest increase in mortality was evident in the second quarter (days 30–35). Populations at the two warmer temperatures (13 and 16°C) displayed high survival and short larval life durations (Fig. 3(c) and (d)). Populations held at 10°C were intermediate in their magnitudes of mortality and in their durations of larval life compared to the warmer and cooler populations (Fig. 3(b)). The age-specific pattern of mortality in the

10°C populations is similar to, though less dramatic than, that observed in the 7°C populations.

All of the survival models detected significant temperature effects on mortality but the models differed in their portrayal of the observed mortality schedules. The exponential-decay and the accelerated-failure-time models described the survivorship curves at the two warmest temperatures reasonably well, but neither model captured the precipitous declines in the
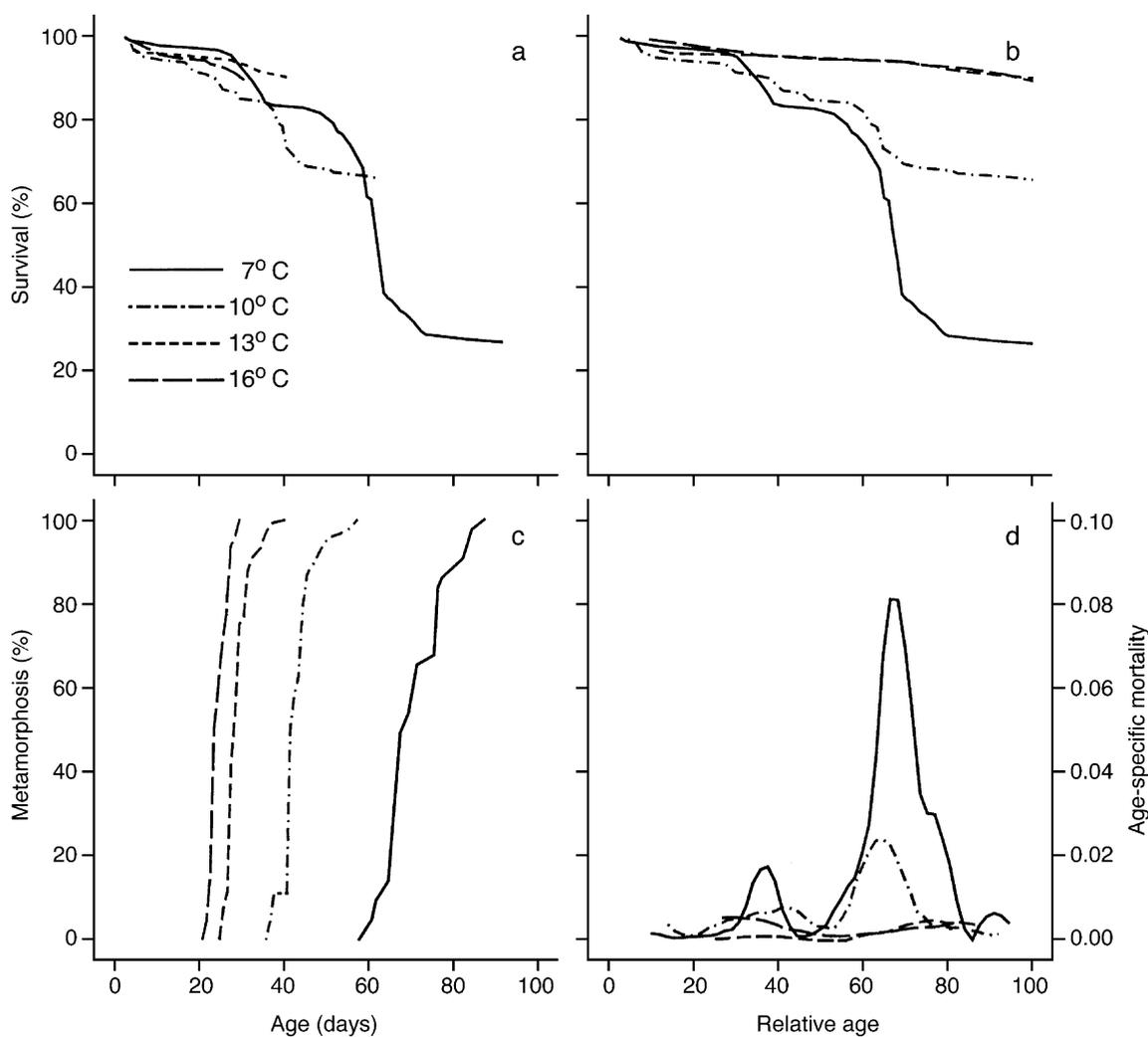
Fig. 5. Predicted survivorship curves, time-course of metamorphosis, and age-specific mortality risks for laboratory populations of larval winter flounder, *Pseudopleuronectes americanus*, reared in the laboratory from hatching through metamorphosis (stage H) at constant temperatures of 7, 10, 13, and 16° C. (a) Predicted survivorship curves from a proportional-hazards model with time-varying covariates. (b) Survivorship curves as in a but with age expressed as relative age (= the proportion of maximum larval life duration at each temperature). (c) Cumulative percentage of all individuals observed to reach metamorphosis (stage H) at each temperature (replicates pooled). (d) Age-specific probabilities of mortality predicted by a proportional-hazards model with time-varying covariates (age expressed as relative age).

7°C populations or the more modest reductions in abundances in the 10°C ones (Fig. 4(a)–(c)). The predicted values from the proportional-hazards model also portrayed the 13 and 16°C populations well. In addition, this model captured some of the observed late-stage increases in mortalities in the two cooler temperature treatments (Fig. 4(d)). The assumption of a consistent and parallel effect of temperature in

the proportional-hazards model, however, resulted in an overestimate of mortality in the second quarter of larval life and an underestimate of it in the third quarter for populations at 7°C. In contrast, the predicted values from the proportional-hazards model with time-varying covariates (Fig. 5(a)) closely match the observed age-specific, environmentally dependent pattern of mortality for all populations (Fig. 4(a)).

The water temperatures experienced by the flounder larvae affected their survival probabilities and durations of larval life (note the temperature-specific lengths of the survivorship curves in Figs. (3) and 4(a)). The termini of the survivorship curves reflect the time at which all larvae had either died or metamorphosed. The time-course of mortality in larval life in general, and the concentration of mortality in the 7 and 10°C populations during the third quarter in particular, are more evident when age-specific mortalities are standardised by the duration of larval life for each temperature. This can be achieved for each temperature by converting age to relative age and computing the proportions of the entire larval life for any age-specific event (Fig. 5(b) and (d)). Using this relative age scale, it is clear that mortalities in the 13 and 16°C populations were low and evenly spaced, whereas mortalities in the 7 and 10°C populations increased moderately in the second quarter of larval life, and increased dramatically during the third quarter (Fig. 5(b) and (d)). The high levels of mortality in the 7 and 10°C populations are concurrent with the metamorphosis of surviving individuals, which is initiated at approximately three-fifths of the maximum duration of larval life for all temperatures (Fig. 5(c)).

4. Discussion

4.1. Evidence of age-specific critical periods

Knowledge of the magnitude, time-course, and sources of mortality in fish populations is fundamental to understanding the large variation in recruitment displayed by many marine fishes. Our investigation shows that it is reasonable to collect data on the magnitude and age specificity of mortality in laboratory populations of marine fish larvae, and that the resulting survivorship curves are repeatable measures of the population status under prevailing environmental conditions. Although the mortality patterns that we report for winter flounder likely depend on the specific rearing environment, including larval diet, we reach the following conditional conclusions. For winter flounder experiencing cool water temperatures (≤10°C), a critical period is expressed during the latter portion of larval life and, to some degree,

during the interval following first feeding but before notochord flexion. This earlier critical period is more likely to be expressed at cooler water temperatures (≤7°C) than those eliciting elevated levels of mortality later in larval life. Winter flounder larvae that experience warmer water temperatures (≥13°C) show no evidence of critical periods and survive at consistently high levels throughout their brief pelagic life. These patterns of age- and environmentally-dependent mortality, and the survivorship curves that represent them, provide a baseline for comparison with other empirically derived or expected survivorship curves.

We detected little if any evidence of a critical period at the time of first feeding of winter flounder larvae and thus conclude that first feeding does not appear to be a period of intrinsic stress for larvae that results in elevated levels of mortality. Using a bioenergetics approach based on experimental data, Laurence (1977) provides simulations of larval winter flounder populations from which he concluded that larvae are likely to experience a critical period at the time of first feeding. The smaller of two peaks in mortality at the coolest temperature in our study (7°C, Fig. 5(d)) occurred after larvae had established feeding. The lack of an increase in mortality at the time of first feeding in our study may be a consequence of our offering an abundance of prey to winter flounder larvae. Based on Laurence's (1977) experimental results and simulations, our rates of feeding of winter flounder larvae (daily additions of five rotifers per cm^3) are $\approx 6\times$ greater than Laurence's calculated threshold prey density, below which first-feeding winter flounder larvae at 8°C would have insufficient hours of daylight to consume subsistence levels of prey. Patterns of larval winter flounder abundances in nature do not resolve whether a critical period occurs at first feeding. Pearcy (1962) reports mortality in larval winter flounder to be at least twofold higher in the first few weeks of larval life than for older larvae. Although this pattern is consistent with a critical period at the time of first feeding, Pearcy's data are based on a catch-curve analysis that used a length-to-age transformation to estimate age, and then separated larvae into two age groups (9–25 days and 26–53 days). We view his data as too highly aggregated to support a conclusive test of the existence of an age-specific critical period at first feeding.

The pattern of mortality beyond the age at first feeding that we report is similar to mortality patterns found in some but not all previous studies of survival in laboratory populations of larval flatfishes. A minimal level of mortality at first feeding and a concentration of mortality later in larval life were reported for California halibut, *Paralichthys californicus* (Gadomski and Caddell, 1991). This contrasts with survivorship curves for larval European plaice, *Pleuronectes platessa*, and sole, *Solea solea*, reared under various conditions over multiple years (reviewed in Shelbourne, 1975). In the latter studies, most deaths occurred during the first half of larval life (between 20 and 30 days of age) with mortality significantly lower and constant thereafter. Even though the magnitude of larval mortality that Shelbourne reports lessened over the years, perhaps due to improved fish husbandry, the time-course of mortality remained similar. Data on mortality at the end of larval life in nature are difficult to find and interpret. Pearcy's (1962) catch-curve data, which are the basis of his estimates of mortality rates for winter flounder larvae, are for larvae that 'showed no signs of metamorphosis' (Pearcy, 1962, p. 25). Pearcy acknowledges that his pelagic gear undersampled settling larvae; consequently, we do not view his data as appropriate for examining the possibility of a critical period concurrent with settlement as we report.

Our findings of temperature effects on the pattern of mortalities (i.e., low survival at cool though viable temperatures, and an interaction between temperature and age-specific mortality) are consistent in part with several prior investigations of flatfishes. Laurence (1975, 1977) found overall survival to metamorphosis of larval winter flounder maintained in the laboratory at 8°C to be as high as 34%, but none survived to metamorphosis at the lowest water temperature evaluated (2°C). Laurence did not report the time-course of mortality. Gadomski and Caddell (1991) report increased mortality of California halibut larvae at the lowest temperature that they evaluated (12°C versus 16, 20, and 24°C), and that mortality was concentrated mid-way through larval life. Burke et al. (1999) also found higher mortality for populations of summer flounder, *Paralichthys dentatus*, experiencing cooler temperatures (18 and 20°C versus 22°C), and that mortality was concentrated around

the time of metamorphosis. In their study, however, cannibalism appeared to be the operative mechanism leading to increased mortality.

Our winter flounder larvae were maintained under food-rich and predator-free conditions and therefore cannot be used to evaluate the patterns of mortality due to starvation or predation in less benign and more natural environments. Several previous studies, however, suggest a nutritional basis for mortality late in larval life that may pertain to our data as well. Early attempts at rearing turbot, *Scophthalmus maximus*, for example, revealed pronounced and repeatable demises in larval numbers at the time that larval prey shifts from rotifers to *Artemia* (Jones et al., 1981). A nutritional limitation may be responsible for the high mortality we observed for winter flounder larvae in their third quarter of pelagic life but, interestingly, this only occurred at the cooler temperatures. There exist numerous examples of fish larvae in starved condition in nature and some studies found starvation to be an important source of mortality for a cohort (Shelbourne, 1957; O'Connell, 1980; Hewitt et al., 1985; Theilacker, 1986; McGurk, 1989). Future experiments on the entirety of larval life that include data protocols such as those reported here, but which also employ a range of prey concentrations for flatfish larvae (Laurence, 1977), would substantially illuminate the relationship between food and age-specific mortality.

4.2. Research recommendations

Our study has raised three concerns regarding the detection and analysis of critical periods in flatfish larvae. First, mixed-aged populations are unlikely to reveal age-specific critical periods unless population members can be aged and monitored at a resolution finer than the expected duration of the critical period. Second, temporal or spatial aggregation of abundance data will likely render these data of little value for resolving critical periods. Third, when abundance-at-age data are available, the statistical model must be capable of capturing irregularities that might signify a critical period. We view these three concerns as sufficient to justify a re-examination of previous research cited as examples of no evidence of critical periods (Marr, 1956; May, 1974; Leggett and Deblois, 1994).

We believe that process-based hypotheses of recruitment variation must ultimately be evaluated with field data. Ideally, these field efforts would be coordinated with laboratory-based validation studies. The following research situations may provide the abundance-at-age data needed for evaluating critical periods in nature. First, if larvae from repeated ichthyoplankton collections could be aged to daily or near-daily resolutions (Yoklavich and Bailey, 1990) researchers would be able to follow abundances of daily cohorts, estimate age-specific losses, and construct survivorship curves. Second, in some natural circumstances larvae are discernible into daily or near-daily cohorts by their location, body size, or the reproductive synchrony of their parents. In such situations, larvae could be tracked, repeatedly sampled, and age-specific mortalities estimated after accounting for other sources of loss of larvae (Taggart and Leggett, 1987; McGurk, 1989; Dorsey et al., 1996). Third, mark-recapture methods applied to large numbers of equal-aged young fish (e.g., just-hatched larvae) could provide estimates of abundances-at-age in nature and thus permit the evaluation of age-specific mortality (Secor et al., 1995). Fourth, like-aged fish could be released into large-scale enclosures and subsequently sampled for abundances-at-age from which age-specific mortality rates could be computed (Moksness and Øiestad, 1987; Wespestad and Moksness, 1989; Blom et al., 1994).

Our study highlights some of the constraints of most traditional field approaches in the quest for detecting critical periods in larval fishes. While acknowledging the limitations of a strictly laboratory approach to the problem, we believe that the experimental examinations we conducted help to identify the type and resolution of data as well as the statistical methods that will aid in the evaluation of patterns of age-specific mortality in nature. Our results also emphasise a potentially important interaction between age-specific mortality and an environmental variable. The presence of such interactions is fundamental to the conceptual linkage between critical periods and year-class variation.

We contend that analysis of critical periods in the early life history of fishes will require data on either ages-at-death or abundances-at-age. Historically, it has been impractical or prohibitively expensive to obtain these data from natural populations of larval fish. The prospective research situations described above, for example, all require high-frequency sampling of cohorts over the appropriate portion of larval life, and one requires otolith-base aging of fish. Furthermore, all assume that changes in abundances-at-age can be apportioned correctly to mortality versus other sources of loss, such as gear avoidance and migration out of the sampled area. With judicious selection of study systems, however, we believe that age-specific mortality of larvae could be estimated, thus yielding a fairer appraisal of critical periods in nature.

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