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Eileen E. Hofmann
Old Dominion University, ehofmann@odu.edu

Eric N. Powell

Eleanor A. Bochenek

John M. Klinck
Old Dominion University, jklinck@odu.edu

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A modelling study of the influence of environment and food supply on survival of *Crassostrea gigas* larvae

Eileen E. Hofmann, Eric N. Powell, Eleanor A. Bochenek, and John M. Klinck

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A biochemically based model was developed to simulate the growth, development, and metamorphosis of larvae of the Pacific oyster (*Crassostrea gigas*). The unique characteristics of the model are that it: (1) defines larvae in terms of their protein, neutral lipid, polar lipid, carbohydrate, and ash content; (2) tracks weight separately from length to follow larval condition; and (3) includes genetic variation in growth efficiency and egg quality to better simulate cohort population dynamics. The model includes parameterizations for filtration, ingestion, and respiration, which determine larval growth rate, and processes controlling larval mortality and metamorphosis. Changes in larval tissue composition occur as the larva grows and in response to the biochemical composition of the food.

Simulations of larval growth indicate that departures of temperature, salinity, or food content from optimum levels reduce larval cohort survival, either because of metabolic constraints that result in death, unsuccessful metamorphosis, or increased predation resulting from increased larval lifespan. Temperatures and salinities near optimal values improve larval survival at low food concentration by increasing ingestion rate or growth efficiency. Also, survival at a given food concentration can vary widely depending on food composition, which determines food quality. The simulations suggest that the ratio of carbohydrate + lipid-to-protein may best describe the overall food quality, with optimal food compositions being characterized by ratios near 1.2 to 1.4 over a range of food concentrations. In contrast, food compositions containing too much or too little protein reduce larval survival, even at saturating food concentrations.

In simulations emphasizing genetic variability within the cohort, larvae with high growth efficiency originating from large eggs out-perform other egg quality–growth efficiency combinations over a wide range of temperature, salinity, and food contents. As a consequence, suboptimal temperature, salinity, or food content compresses genetic variation by uniformly favouring larvae from large eggs with a high growth efficiency. However, the larval survival obtained from simulations that use a range of food qualities is representative of a much broader range of genetic types. Thus, the simulations support the supposition that food quality is an important variable controlling the survival and genetic variability of *C. gigas* larval cohorts.

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E. E. Hofmann and J. M. Klinck: Center for Coastal Physical Oceanography, Crittenton Hall, Old Dominion University, Norfolk, VA 23529, USA. E. N. Powell and E. A. Bochenek: Haskin Shellfish Research Laboratory, Rutgers University, 6959 Miller Avenue, Port Norris, NJ 08349, USA. Correspondence to E. E. Hofmann: tel: +1 757 683 5334; fax: +1 757 683 5550; e-mail: hofmann@ccpo.odu.edu.

Introduction

Laboratory and hatchery observations (Dupuy *et al.*, 1977; Gallager *et al.*, 1986; Gallager and Mann, 1986; Quayle, 1988; Robinson, 1992), high year-to-year variability in recruitment (Loosanoff, 1966; Hofstetter, 1977; Austin *et al.*, 1996), and the limited evidence for a broodstock–recruitment relationship (Prytherch, 1929; Peterson and Summerson, 1992; Livingston *et al.*, 2000) suggest that the highly variable survivorship of bivalve larvae in nature is determined by complex interactions among physiology, environmental conditions, and food supply. Variability in the environment or food supply affects survivorship by not permitting completion of larval life or successful metamorphosis or by producing slow growth, thereby increasing larval lifespan and exposure to predators. Considerable research has been conducted on the general topic of larval survivorship under laboratory and hatchery conditions (Helm *et al.*, 1973; Gallager *et al.*, 1986; His and Seaman, 1992; Haws *et al.*, 1993; Laing, 1995), with food quantity and quality receiving particular attention (Helm *et al.*, 1973; Wikfors *et al.*, 1984; Utting, 1986; Thompson and

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Harrison, 1992; Thompson *et al.*, 1994, 1996). Application of these results to study larval survival in the field, however, has been extremely difficult because tracking individual larval cohorts is labour intensive at best and essentially intractable in most environments. Consequently, observations of temporal variation in larval abundance or benthic settlement are often explained by inference (Resnik, 1991). Thus, larval growth (Rice *et al.*, 1993; Dekshenieks *et al.*, 1993, 1997, 2000) and transport models (Jackson, 1986; Hill, 1991; Dekshenieks *et al.*, 1996) provide a framework for examining the role of environmental factors in controlling larval survivorship.

The existing models, however, do not resolve larval physiology and growth at the level of basic biochemical metabolism and composition. As a result, food quantity effects on larval survival have been addressed incompletely and food quality effects have not been examined, in spite of studies suggesting that both are important for shellfish larval survival (see previous references) and the recognition of a critical feeding period for larvae (Lasker, 1975; Cushing and Dickson, 1976; Anger *et al.*, 1981; Cushing 1990, 1995). In this study, a biochemically based larval growth model is used to investigate interacting effects of temperature, salinity, food quantity, and food quality on larval success. The model used was developed for larvae of the Pacific oyster, *Crassostrea gigas* (Bochenek *et al.*, 2001; Powell *et al.*, 2002), and it allows explicit examination of environmental, food quantity, and food quality effects on bivalve larvae survivorship.

The following section provides a brief description of the *C. gigas* larval growth model and of some model parameterizations. This is followed by simulations that illustrate the effects of temperature, salinity, food quantity, and food quality on larval survival and success at metamorphosis. The discussion examines how food quantity and food quality interact with environmental conditions to influence larval survivorship and evaluates the relative significance of food quality as a factor determining larval cohort survivorship.

Model description

Model structure and governing equations

The change in length for an individual larva over time is given by:

$$\frac{dL}{dt} = \alpha L \tag{1}$$

where L is larval length in μm and α is the rate at which the larva grows in units of d^{-1}. Larval growth rate (α) is based on formulations that allow differential metabolism of the protein, carbohydrate, and lipid content of the food ingested by the larva. Thus, net production is expressed as the difference between assimilated ingestion (AI) and respiration (R):

$$NP_i = AI_i - R_i \tag{2}$$

where i represents the four basic biochemical components included in the model: protein, polar lipid, neutral lipid, and carbohydrate. Excretion is assumed to be a minor carbon loss in comparison to respiration. The model given by Equations (1) and (2) is an individual based model and, as such, represents the average individual in a population. The simulations obtained from the individual based model are extended to include the genetic variation of the larger larval population, as described subsequently.

An increase in larval length occurs when the sum of the four biochemical components that contribute to net production, $\sum_{n=1}^{4} NP$, is positive, when larval condition index is maximal for a given size, and when the restrictions imposed by certain biochemical ratios, which are based on molecular weight and described subsequently, are simultaneously met. Thus, excess net production, ENP, is the basic quantity responsible for larval growth and this determines α in Equation (1). The value of ENP is determined from filtration rate, the metabolism of carbohydrate, polar lipid, neutral lipid, and protein, and the conversion of the metabolized food into structural components and storage material. A basic assumption in this model is that the formation of structural components determines larval length increase. Material converted into storage components, i.e. neutral lipids, does not result in growth in length. The conversions and parameterizations used to model these processes are described briefly in the following sections, and details are given in Bochenek *et al.* (2001) and Powell *et al.* (2002).

Assimilated ingestion in Equation (2) is expressed as the product of filtration rate (FR), ingestion efficiency (IE and IE_s), temperature and salinity (TS factor), food ($Food_i$), assimilation efficiency (AE_i), and a small larvae factor (SLF_i) as:

$$AI_i = FR \times IE \times TS\ factor \times Food_i \times AE_i \times SLF_i \tag{3}$$

where IE is replaced by IE_s for larvae smaller than 80 μm. Details of the parameterizations used for the processes included in Equation (3) are given in Bochenek *et al.* (2001) and Powell *et al.* (2002).

The respiration rate for *C. gigas* larvae is parameterized from data presented in Gerdes (1983) and Hoegh-Guldberg and Manahan (1995) as:

$$Resp = r_0 W^{\theta} \tag{4}$$

where Resp is given in ml O_2 consumed $ind.^{-1}\ h^{-1}$ and W_i is dry tissue weight in mg. The coefficient θ has the value of 0.95 and the base respiration rate, r_0 in kJ d^{-1}, is specified using a distribution (described in a following section) that is assumed to reflect genetic variations in metabolic processes that are known to occur for individual *C. gigas* larvae (e.g. Lannan, 1980). The respiration rate obtained from Equation (4) is apportioned among the four biochemical components to determine the respiration portion of net production given by Equation (2).

The energy needed to cover the respiration rate obtained from Equation (4) is first taken from the assimilated

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carbohydrate pool (Table 1). If this pool is insufficient to meet the cost of respiration then the remaining deficit is taken from the neutral lipid pool and any remaining deficit is then taken proportionately from the structural components of the larva (Table 1).

The *C. gigas* larval model given by Equation (1) was solved numerically using a third-order Adams-Bashforth scheme (Canuto *et al.*, 1988) with a time-step of 0.1 day, which is sufficient to avoid numerical diffusion during the larval lifespan, which averages about 2 to 3 weeks depending on environmental conditions.

Parameterization of processes

Preferred biochemical composition

Certain weight ratios between structural constituents are assumed to be associated with healthy larvae. These ratios are used to allocate assimilated food constituents, when in sufficient supply, to tissue pools. Deviations in the resulting tissue composition from these ratios result in larval mortality. The relationship between tissue lipid and protein was obtained from His and Maurer (1988) under the assumption that the total lipid content of *C. gigas* larvae, like *C. virginica* larvae, is about evenly split between neutral and polar lipids (Gallager *et al.*, 1986; Whyte *et al.*, 1987). This yielded a preferred tissue polar lipid-to-tissue protein ratio of 0.11 for healthy larvae. The equivalent ratio between tissue carbohydrate and tissue protein was determined to be 0.01 (Gallager *et al.*, 1986; Whyte *et al.*, 1987; His and Maurer, 1988). Both ratios are independent of larval size. Most tissue carbohydrate was assumed to be structural, because neutral lipid is the primary storage constituent.

Fate of assimilated ingestion

The fate of the assimilated protein, neutral lipid, polar lipid, and carbohydrate differs within the larva (Table 1). Protein assimilated in a given time interval has the somatic protein pool as its primary destination. Protein may also be used to cover a respiratory deficit in accordance with the appropriate protein:carbohydrate:polar lipid ratio.

The carbohydrate needs of the larva are determined by the amount required to maintain somatic carbohydrate in its proper proportion and that needed to cover the cost of respiration (Table 1). The amount of somatic carbohydrate needed to maintain the carbohydrate:protein ratio (0.01) is debited from the available assimilated carbohydrate and added to the carbohydrate pool, with the excess becoming part of the larval neutral lipid reserve. When tissue imbalances occur, e.g. insufficient polar lipid to meet the tissue compositional requirements of the larvae, somatic carbohydrate is used to maintain larval polar lipid in its proper proportion. Assimilated carbohydrate is the primary means by which larval respiratory needs are met (Table 1).

Assimilated polar lipid is added to the somatic polar lipid pool in accordance with the protein:polar lipid ratio (Table 1), with the excess going to the neutral lipid pool. When carbohydrate imbalances occur, polar lipid reserves are mobilized to produce somatic carbohydrate in an amount that is consistent with maintaining the protein:carbohydrate ratio. Polar lipids are also used to cover deficits arising from respiratory demands.

The primary destination of assimilated neutral lipid is the neutral lipid pool (Table 1). This internal pool is mobilized to maintain somatic carbohydrate and somatic polar lipid pools in accordance with the appropriate ratios, when assimilated carbohydrate and polar lipid are not present in the proper proportions in the food. The neutral lipid pool can also be used to cover respiratory needs during periods of carbohydrate deficit. This pool also provides a means for larvae less than 80 μm to produce somatic carbohydrate, polar lipid, and protein as well as cover respiratory costs early in larval life. At any point in the development of the larva, the inability to maintain one of the biochemical constituent ratios, or the inability to remove a deficit in a biochemical pool results in death of the larva.

Table 1. Destination of assimilated protein, carbohydrate, polar lipid and neutral lipid in *Crassostrea gigas* larvae. The particular biochemical ratio determining the conversion to individual reservoirs is indicated in brackets. Columns 2, 3, and 4 indicate the fate of the food; column 5 indicates the fate of the tissue. Transfers of food that do not occur in response to deficit or surplus conditions are indicated as not applicable (NA). Protein, carbohydrate, and polar lipid are indicated by P, C, and PL, respectively.

Food constituent	Primary destination in larva	Food deficit response	Food surplus response	Tissue maintenance deficit	Early life (<80 μm)
Protein	Somatic P	NA	NA	Respiration [P:C:PL]	NA
Carbohydrate	Somatic C and respiration [P:C]	Somatic PL [P:PL]	Neutral lipid reserve	Respiration [P:C:PL]	NA
Polar lipid	Somatic PL [P:PL]	Somatic C [P:C]	Neutral lipid reserve	Respiration [P:C:PL]	NA
Neutral lipid	Neutral lipid reserves	Somatic C [P:C] Somatic PL [P:PL] Respiration	NA	Somatic C [P:C] Somatic PL [P:PL] Respiration	Somatic C Somatic PL Somatic P Respiration

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Larval growth

Larval growth in a given time interval is based on maintaining the protein:ash ratio for a given larval length, as described in Bochenek *et al.* (2001). An increase in larval length occurs when the protein, carbohydrate, and polar lipid pools are in excess of what is needed to maintain the protein:ash ratio at a given length, which defines ENP. During times of protein deficit with respect to ash weight (low condition index), the larva can have a positive net production that increases organic mass and condition index, but produces no excess net production and, hence, no increase in length. Periods during which the larva resorts to using structural material to cover respiration costs result in reduction of larval condition index, which is manifest as a reduction in the protein-to-ash ratio, because the somatic tissue pool shrinks with respect to shell weight.

Larval metamorphosis

Observations suggest that *C. gigas* larvae may initiate metamorphosis once they reach 275 μm and that this process may or may not be successful (Ventilla, 1984; Kusaki, 1991; Laing, 1995). Thus, the simulated larva is assumed to have the potential of becoming competent for metamorphosis at 275 μm. Metamorphosis is triggered when the larva experiences a 25% drop in neutral lipid stores in one day, a rate that, if continued, impairs successful metamorphosis. Once competent, the larva immediately attempts metamorphosis and success occurs if the larval neutral lipid pool is greater than the polar lipid pool. This establishes a minimum storage requirement needed to sustain metamorphosis. If this condition is not satisfied, then metamorphosis is unsuccessful and the larva dies. Further justification for this modelling approach is given by Powell *et al.* (2002) and Bochenek *et al.* (2001).

The process of larval searching for appropriate substrate (e.g. Hidu and Haskin, 1971; Roegner and Mann, 1990; Ólafsson *et al.*, 1994) is not included in the model. Successful larvae in the model are assumed to have sufficient reserves to sustain such a search. Varying searching times in the model would simply add a random factor to the success of any larval genotype and would not advance the primary goal of the model, which is to examine the influence of food quantity and food quality during larval life on larval cohort survival. Thus, an implicit assumption in the model is that substrate is not a factor limiting survivorship, or, if so, that its influence is not a function of the larval energy budget.

Biochemically determined metabolic mortality

The simulated larval growth prior to metamorphosis is based on maintaining specific ratios between protein, polar lipid, carbohydrate, and ash weight. Small variations in these ratios, which are consistent with changes that occur in the larva as it grows, are allowed. However, large changes are not permissible. The inter-dependencies of the biochemical ratios result in the protein:ash ratio being a good indicator of the biochemical state of the larva. If this ratio is reduced at any time to 70% or less of its needed value, then the larva is assumed to die (Bochenek *et al.*, 2001). This condition is termed starvation in the model.

During the initial stages of larval growth, about the first two days, the larva does not filter efficiently and hence food ingestion is not usually sufficient to cover metabolic costs. During this period it is assumed that the larva survives by using its stored neutral lipid supply. However, if during this period the neutral lipid supply approaches zero, the larva is assumed to have reached its metabolic point of no return and dies. Also, inability of the larva to maintain its required protein:lipid or protein:carbohydrate ratios results in death.

Model implementation and analysis

Initial C. gigas *egg size, including genetic variability*

The eggs spawned by *C. gigas* adults have an average size of 50 μm (Quayle, 1988; Arakawa, 1990). However, using this as the initial condition for the model resulted in mismatches in the initial simulated and observed length-to-weight relationships, which are based on larval size. Thus, simple egg diameter is not the appropriate measure to use with the length-to-weight relationship and other conversions. The discrepancy arises because of the mismatch between the volume of a spherical egg and the more ellipsoidal-shaped larva. Therefore, egg diameter was converted to an equivalent larval size using a diameter-to-length conversion factor of 1.096 (Arakawa, 1990). Thus, a 50-μm egg is equivalent to a 54.8-μm larva.

C. virginica egg size ranges between 30 and 80 μm (Gallager *et al.*, 1986). More limited information is available for *C. gigas*, but a similar range of egg sizes can be inferred. Therefore, the initial conditions for each simulation included egg sizes over this range. This variation was assumed to represent genetically or environmentally determined variability in the spawning population.

To establish the initial biochemical composition of the egg, the larval size immediately post-hatch was used with a *C. gigas* length-to-dry tissue weight relationship (given in Bochenek *et al.*, 2001) to calculate an initial dry weight, which in turn was used to obtain an initial ash weight value (given in Bochenek *et al.*, 2001). The protein component of the egg was then determined by multiplying the ash weight by the protein:ash ratio. The egg polar lipid content was determined by multiplying the protein content by the polar lipid:protein ratio. The carbohydrate content was taken to be 1% of egg dry weight. Neutral lipid content was obtained by difference. A negative value for neutral lipid was assumed to indicate a non-viable egg.

Genetic effects on larval mortality

Certain combinations of initial egg size and respiration rate are assumed to be less common in the larval cohort, and other combinations are less viable overall due to metabolic imbalances, metabolic inefficiencies, or longer larval lifespans, which increase predation loss. Respiration rate is

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used as a proxy for growth efficiency, with efficiency increasing with decreasing respiration rate and vice versa. This type of genetically determined variability within a cohort, GE, is prescribed with a Gaussian function of the form:

$$GE = e^{-(ES-ES_0/2sd_{egg})^2} - e^{-(Resp-Resp_0/2sd_{resp})^2} \quad (5)$$

where the Gaussian distributions extend for 2 s.d. ($2sd_{egg}$, $2sd_{resp}$) about a central egg size of 50 μm, and central respiration rate of 1.046 kJ d^{-1}, given by ES_0 and $Resp_0$, respectively. Equation (5) weights mortality or any other population process by a population distribution that is characterized by a certain range of egg sizes and respiration rates. Thus, the surviving larval population represents the combined effects of genetics, food composition, and environmental conditions.

Extrinsic sources of larval mortality

Termination of a simulation occurs because of successful metamorphosis, unsuccessful metamorphosis, inappropriate metabolic ratios, and starvation. The larval size and total development time for each combination of initial egg size and respiration rate are output as diagnostics when the simulations end. These diagnostics are evaluated to determine the potential effect of extrinsic mortality, which arises primarily from predation, on larval survival. The extrinsic mortality (EM) is assumed to increase in proportion to the larval lifespan associated with each combination of egg size and respiration rate, and is imposed with a relationship of the form:

$$EM(j,k) = e^{-m_0 LD(j,k)} \quad (6)$$

The time required for a larva with an initial egg size (j) and respiration rate (k) to trigger a mortality event or to successfully metamorphose, LD, is obtained from the larval growth simulations. The daily mortality rate, m_0, has the value 0.143 d^{-1}, which is the same as that used by Dekshenieks *et al.* (1997) to specify mortality for *C. virginica* larvae. Thus, the resultant extrinsic mortality, which is calculated separately from the growth simulation, provides an additional filter for larval survival.

Food inputs

Most of the larval growth simulations were done using a food quantity of 2 mg L^{-1}, and a food quality defined by a ration consisting of 46% protein, 39% carbohydrate, 9% polar lipid, and 6% neutral lipid, which gives a ratio among the biochemical components of 3:2.5:0.6:0.4. This food composition yields an assimilation efficiency near 70% and is referred to as 'standard' food. These food conditions were held constant over the course of larval life.

Additional simulations used a range of food qualities that were specified based on carbohydrate, lipid, and protein content of selected algal species reported in Lee *et al.* (1971), Roman (1983), Parsons *et al.* (1984), Wikfors *et al.* (1984), and Utting (1986). The measured compositions of taxa such as *Tetraselmis*, *Dunaliella*, *Amphidinium*, *Monochrysis*, *Thalassiosira*, and *Isochrysis* were used with the model rather than artificially created sequences to increase the realism of the simulations. The influence of food quality on survival was then assessed through the use of ratios between the primary constituents of these algal types: protein-to-lipid, carbohydrate-to-lipid, and carbohydrate-to-protein.

Presentation of simulation results

The simulation results are analysed in terms of three metrics. The first metric focuses on the degree to which different genotypes (e.g. large eggs, low growth efficiency vs. big eggs, high growth efficiency) are intrinsically selected for by different environmental conditions. All extrinsic factors, such as predation, are excluded as sources of mortality in this metric in order to focus on inherent genetic capabilities. The second is total cohort survival through metamorphosis. This metric, integrated across all egg sizes and respiration rates as determined by the Gaussian distribution of genetic traits in the cohort, given by Equation (5), describes the sum of all intrinsic and extrinsic sources of mortality. It is sensitive to physiological constraints as well as to cumulative predation pressure controlled by variations in larval lifespan. Thus, as larval lifespan increases, total cohort mortality increases, because the exposure period to predation increases. The third metric, the ratio between total survival and intrinsic survival, provides a mechanism by which to assess the importance of predation in total survivorship. The ratio is not defined for environmental conditions that do not permit any larvae to survive through metamorphosis (100% intrinsic mortality).

Results

Basic simulation

The simulated fate of *C. gigas* larvae differs considerably over the range of initial egg sizes and respiration rates used to represent cohort variability (Figure 1). An obvious result is that larvae that begin life as eggs that are between 30 and 37 μm do not survive at any respiration rate. These larvae have initial biochemical ratios that result in non-viability. Large eggs, those between 74 and 80 μm, are also non-viable because of a metabolic constraint at day 2 which produces insufficient neutral lipid to continue development. Similarly, larvae at small initial egg size, with respiration rates greater than 0.9 kJ d^{-1}, experience the same metabolic constraint on continued development. There is a wide range of initial egg sizes and respiration rates that allows larvae to develop to the point of metamorphosis. However, upon reaching metamorphosis, the biochemical constituents of many larvae are not in the appropriate ratios to sustain successful metamorphosis. Development through successful metamorphosis occurs for 78% of the initial egg sizes, but only for about 36% of the respiration rates. Thus,

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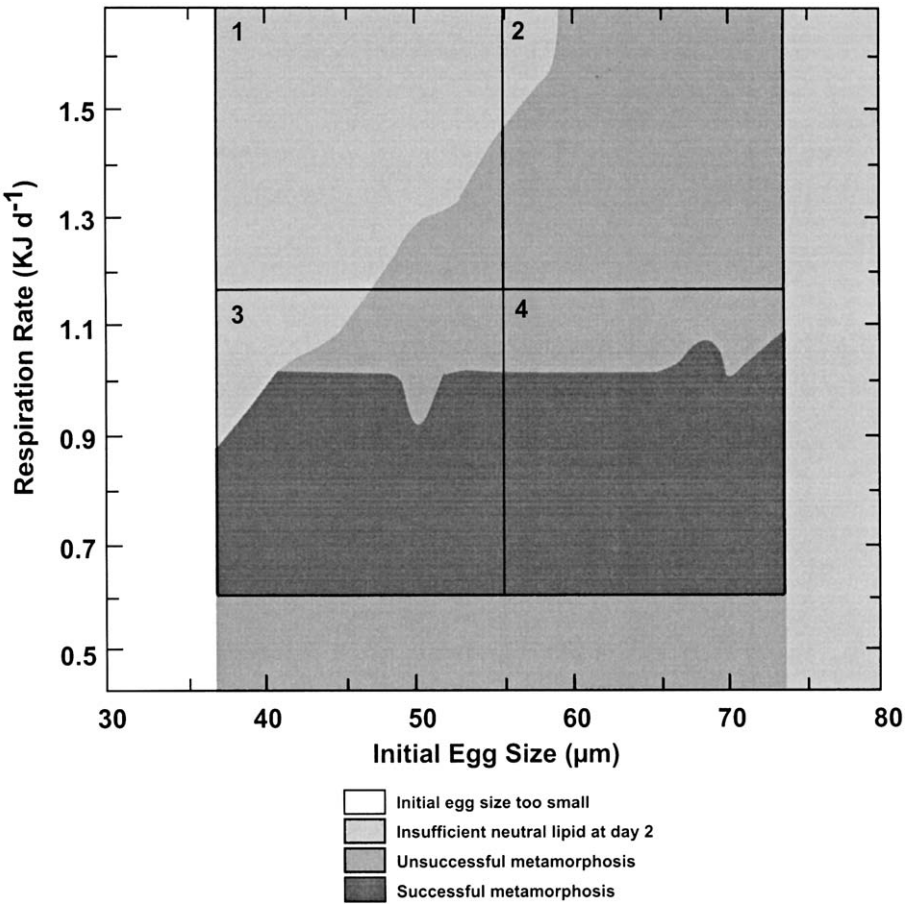

Figure 1. Simulated fate of *Crassostrea gigas* larvae for a range of initial egg sizes and respiration rates exposed to a standard food ration of 2 mg L^{-1} at 27.5°C and 17.5 psu. The boundaries of the four quadrants used to calculate survival factors for various genetic compositions are marked. Survival factors calculated for this simulation are, for quadrants 1, 2, 3, and 4, respectively, 1.15, 1.9, 2.5, and 2.7.

respiration rate (growth efficiency) is a stronger determinant of larval survival than is initial egg size for the conditions used in this simulation.

To further quantify the effect of initial egg size and respiration rate on larval success, the two-dimensional array describing genetic variability was divided into four quadrants (Figure 1) and a survival factor for each computed. The extremes that rarely yield viable larvae were excluded from the analysis. Within each quadrant, the individual genotype combinations that die prior to metamorphosis are assigned a survival factor of 1, those that survive to metamorphosis but fail to complete metamorphosis are assigned a survival factor of 2, and those that undergo successful metamorphosis have a survival factor of 3. The mean of the survival factors assigned to the individual genotypes gives an average survival for each quadrant. For example, if there are three larvae and one fails to undergo metamorphosis and two survive, the corresponding survival factor is 2.67 (2 + 3 + 3/3).

The mean survival factor for quadrant 1 (Figure 1) is 1.5, which indicates that the majority of these genotypes die prior to metamorphosis. In contrast, the average survival factors for quadrants 3 and 4 are 2.5 and 2.7, respectively, indicating that most of these genotype combinations successfully complete metamorphosis. The average survival factor for quadrant 2 is 1.9, which shows that these genotypes survive to attempt metamorphosis, but are unsuccessful in completing the process.

Temperature and salinity

When food was plentiful, significant cohort survival (defined as $\geq$2% survival through metamorphosis, second metric) occurred over a broad salinity range as long as temperatures remained above 24°C (Figure 2A). More than 10% of the cohort survives through metamorphosis at temperatures of 27.5° to 28°C and salinities of 25 to 27.5 psu. Larval survivorship declines to near zero below 23°C, regardless of salinity. In comparison, even at the extremes of salinity, 20 and 35 psu, larval survivorship rarely falls below 2% when temperature exceeds 24°C.

At 1.0 mg L^{-1} food, cohort survival reaches 2% at 24°C only for salinities near 25 psu (Figure 2B). At 0.75 mg L^{-1},

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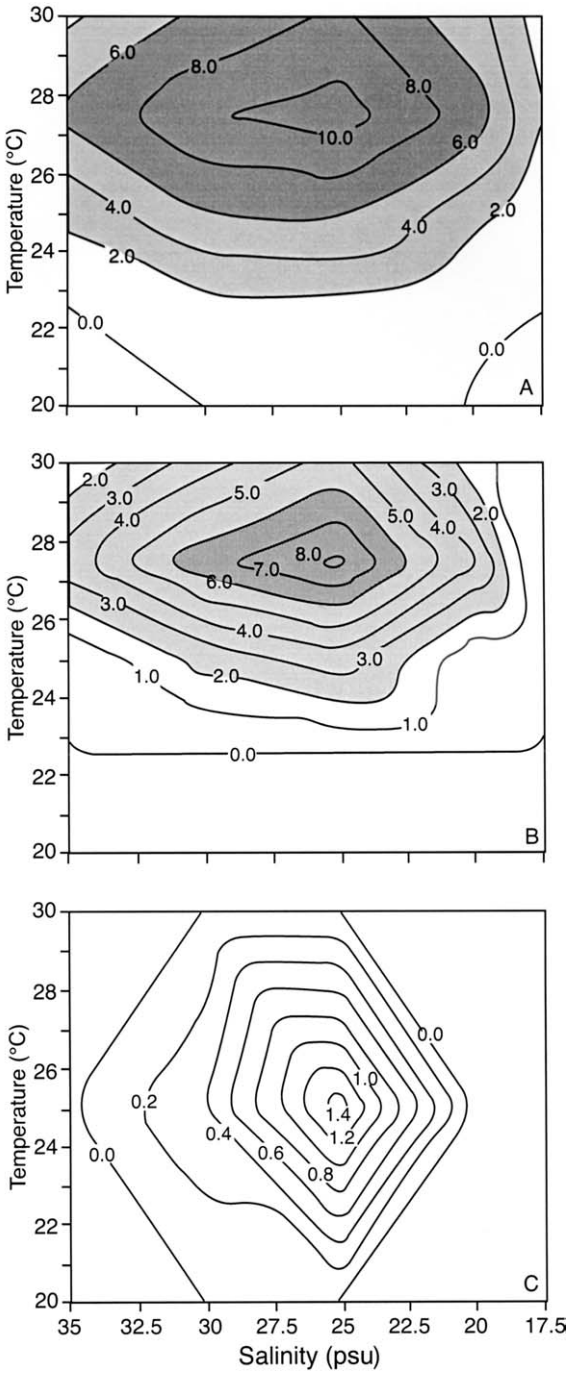

Figure 2. Simulated temperature and salinity dependence of the percent of total cohort survival through metamorphosis using the standard food quality provided in a ration of (A) 1.5 mg L^{-1}, (B) 1.0 mg L^{-1}, and (C) 0.75 mg L^{-1}. Note that the x-axis goes from high to low salinity. Cohort survival of 2–6% and ≥6% is indicated by the light and dark shading, respectively.

cohort survival never exceeds 1.4% for any temperature–salinity combination (Figure 2C). Thus, lower food rations compress the range of temperatures producing significant cohort survival. Similarly, lower food rations compress the range of salinities producing significant cohort survival. For a ration of 0.75 mg L^{-1}, cohort survival rises above 1% only for salinities of 22.5–27.5 psu.

Not all genotypes are equivalently successful over different temperature–salinity combinations. At a food ration of 2.0 mg L^{-1}, larvae with high respiration rates (low growth efficiencies) rarely metamorphosed successfully (Figure 3A); however, larger eggs had a higher success rate (Figure 3B). The survival factors rarely exceeded 2 for small eggs, but exceeded 2 for large eggs over a wide salinity range at temperatures above 24 °C. Larvae with low respiration rates (high growth efficiencies) reached a larval survival factor of 2.5 over a broad salinity range at most temperatures above 24 °C for small and large initial egg sizes (Figure 3C, D). Most of these larvae successfully metamorphosed (survival factor ≥2.5), especially those that began life as large eggs (Figure 3D).

A 50% reduction in food supply compresses the range of temperature and salinity that favours larval survival (Figure 4A, B vs. Figure 3A, B). Failure to survive to metamorphosis is increased, particularly for larvae from small eggs with low growth efficiencies (Figure 4A). Larvae coming from large eggs with high growth efficiencies have the greatest survival (Figure 4B).

Food quantity, temperature, and salinity

Food supply and temperature interact to produce a range of variability in larval cohort survival (Figure 5). At a given salinity, survival is greatest at 27.5 °C over a range of food rations (Figure 5A, B), although it is less at lower salinity. Survival increases to a food ration of about 1.5 mg L^{-1}, after which no additional increase occurs. At 25 psu, significant survival (>2%) occurs at most food rations above 0.8 mg L^{-1} above 22.5 °C (Figure 5A). Below a food value of 1.5 mg L^{-1} at 20 psu (Figure 5B) growth is reduced, which increases larval lifespan and loss to predation. Reduced salinity compresses the range of optimality to temperatures above 25 °C and food rations above 1 mg L^{-1}. Increased food supply permits survival at lower temperatures at optimal salinity (Figure 5A), but has a smaller effect at lower salinity (Figure 5B).

Genotype survival varies considerably as a function of temperature and food supply (Figure 6). At 25 psu, larvae with high respiration rates (low growth efficiency) rarely metamorphosed successfully at any food supply or temperature combination (Figure 6A, B). However, an initial larger egg size partially offsets the deleterious effect of a higher respiration rate and provides a greater chance for successful metamorphosis, as suggested by a survival factor greater than 2.5, at food rations in excess of 1.3 mg L^{-1} at optimal temperatures (Figure 6B). Larvae with low

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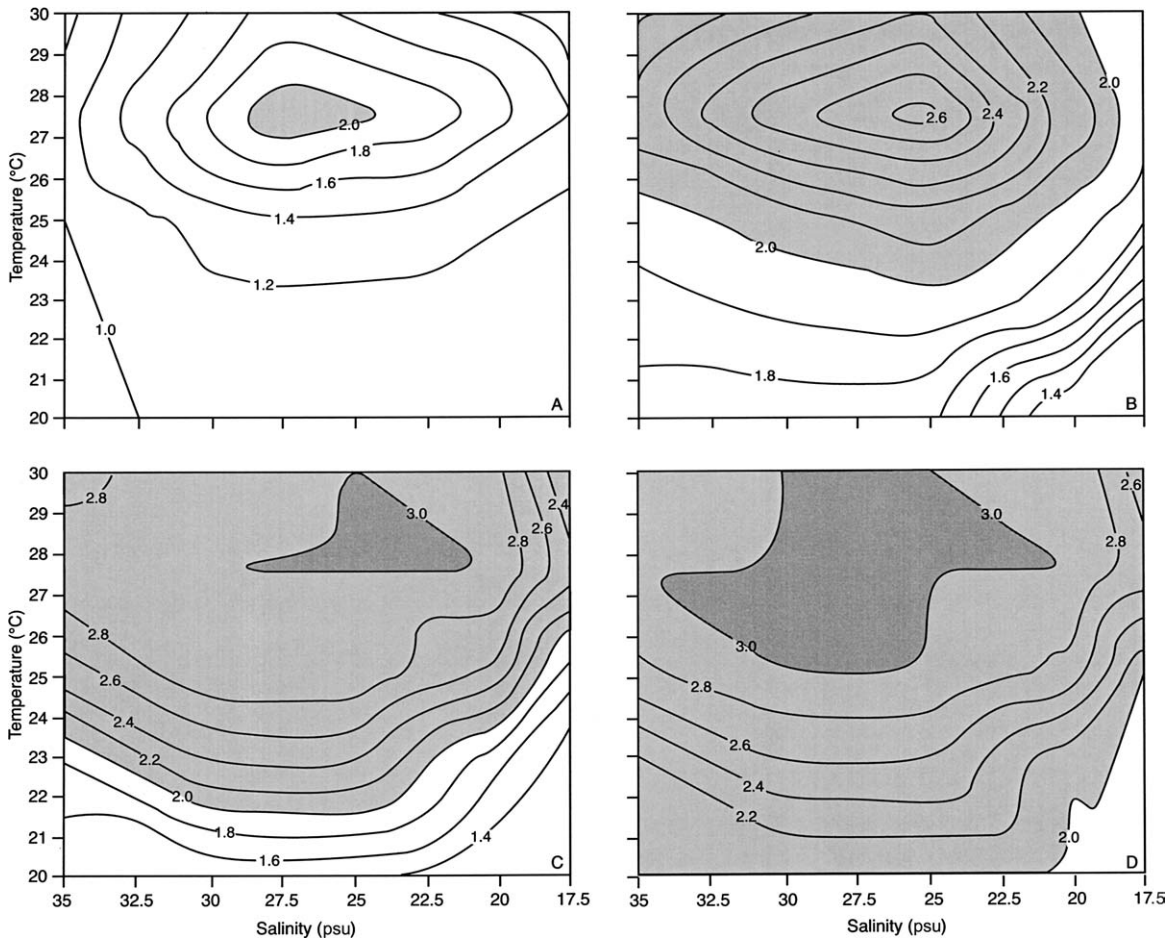

Figure 3. Average survival factor for (A) small eggs with high respiration rates, (B) large eggs with high respiration rates, (C) small eggs with low respiration rates, and (D) large eggs with low respiration rates, obtained from simulations done for a range of temperature and salinity combinations and a standard food ration of 2.0 mg L^{-1}. Contour values are: 1.0–2.0 (unshaded) genotype dies before metamorphosis; 2.0–3.0 (light shading) genotype attempts metamorphosis and some successfully complete the process and others are unsuccessful; and, 3.0 (dark shading) genotype successfully completes metamorphosis.

respiration rates (high growth efficiencies) were able to successfully complete metamorphosis (survival factor of 3.0) over a broad temperature range at most rations above 1 mg L^{-1} (Figure 6C, D). This is most pronounced for large initial egg size (Figure 6D), but genotypes arising from small initial egg size also contribute to the cohort (Figure 6C).

Increased food supply increases larval survival at all salinities up to a ration of 1.5 mg L^{-1} (Figure 7), at 27.5°C. Below food values of 1 mg L^{-1} salinity has only a limited effect on larval survival. At higher food rations, larval survival decreases substantially as salinity diverges from 25–27.5 psu. Thus, at low food concentrations small changes in salinity cannot compensate for reduced food supply.

Simulations suggest that, unlike temperature, the influence of egg size is of little consequence in determining larval survival over a wide range of salinity–food ration combinations (Figure 8). At 27.5°C, larvae with high respiration rates (low growth efficiency) rarely exceed a survival factor of 2.5 at any food–salinity combination, regardless of initial egg size (Figure 8A, B). For these larvae, salinities that diverge from 25–30 psu and food rations below 1.5 mg L^{-1} produce survival factors below 2.5, indicating that over half of the genetic combinations fail to successfully metamorphose, even without extrinsic sources of mortality. Larvae with low respiration rate (high growth efficiency) attain survival factors above 2.8 over most food–salinity combinations at a ration above 1.0 mg L^{-1}, regardless of initial egg size (Figure 8C, D).

Simulations that used lower temperatures (not shown) indicate that the range of genotypes yielding larvae that metamorphose successfully is considerably restricted. Successful metamorphosis only occurs at 25–30 psu at 22.5°C, for example, and successful genotypes are

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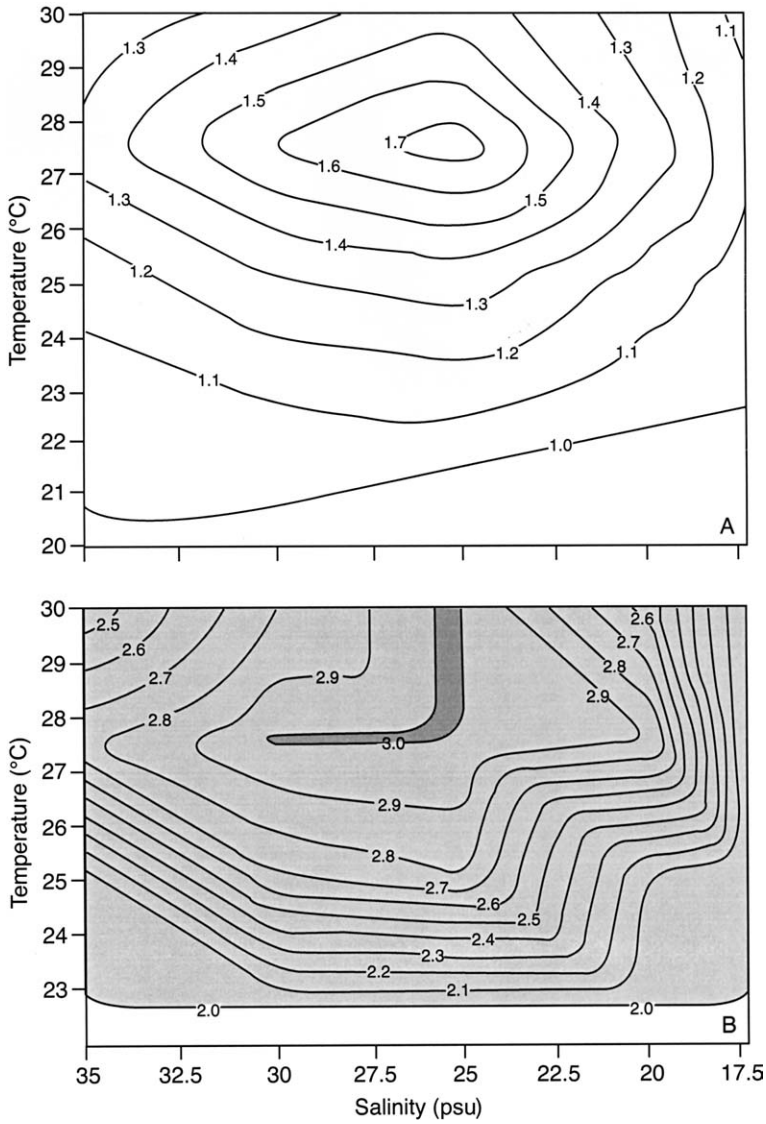

Figure 4. Average survival factor for (A) small eggs with high respiration rates and (B) large eggs with low respiration rates, obtained from simulations that used a range of temperature and salinity combinations and a standard food ration of 1.0 mg L^{-1}. Contour values and shading are the same as in Figure 3.

restricted, for the most part, to larvae coming from large eggs and characterized by low respiration rate (high growth efficiency). No food ration produces successful metamorphosis for larvae with low growth efficiency.

Food quality and food quantity

The percent larval survival was greatest at protein-to-lipid ratios near 3 at high food concentrations; however, optimal ratios for survival declined to 2 or less at low food concentrations (Figure 9A). The larval survival pattern was more complex for carbohydrate-to-lipid ratios, being highest at ratios near 2.5 for food concentrations above about 1 mg L^{-1}, with a broad range of ratios producing survival above 4% (Figure 9B). As food declined below 1 mg L^{-1}, the carbohydrate-to-lipid ratio that retained a survivorship of 2% or greater declined below 2 (Figure 9B). Variation in the carbohydrate-to-protein ratio produced the most complex pattern, with the best survival at ratios near 0.8 and a secondary mode near 0.6 (Figure 9C). Distinctly lower survivorship at a ratio near 0.7 was produced by an algal composition with a protein-to-lipid ratio that limited survivorship, rather than an inherent limitation imposed by a carbohydrate-to-protein ratio near this value. The ratios yielding the greatest survivorships remained relatively stable over a wide range of food contents, although

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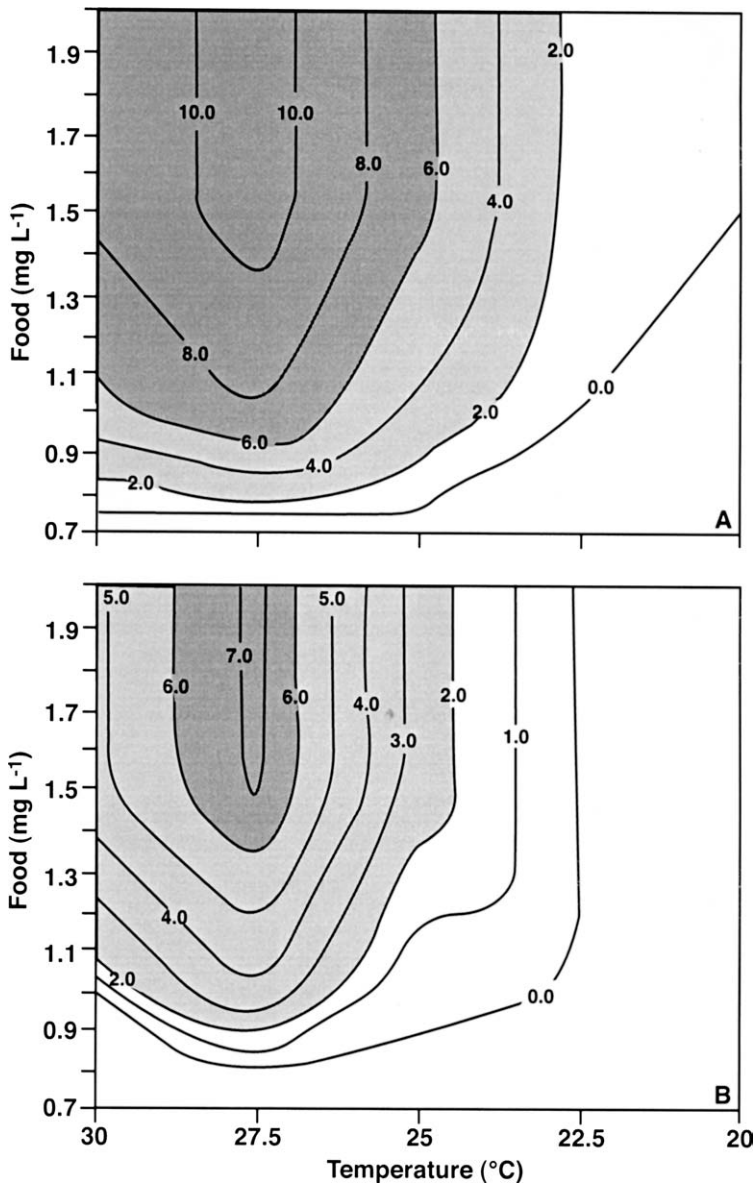

Figure 5. Percent of total cohort survival obtained from simulations that used a range of temperature and standard food ration combinations and salinities of (A) 25 psu and (B) 20 psu. Shading is the same as in Figure 2.

the anticipated decline in survivorship with declining food content did occur (Figure 9C).

Carbohydrate and lipid, though different in many ways, are interchangeable in meeting many of the basic demands of tissue maintenance and energy storage (Table 1). Accordingly, the ratio of protein to the sum of the other constituents is a basic compositional descriptor. The percent larval survival was higher with slightly more lipid + carbohydrate than protein; however, survival was high over a wide range of carbohydrate + lipid contents (Figure 9D). Survival declined precipitously when protein content exceeded carbohydrate + lipid by more than about 0.1 (a ratio of 0.9), and at even higher ratios (near 1.2) when at rations below 1 mg L^{-1} (Figure 9D).

At the extremes, such as a protein-to-lipid ratio much above 4 (cf. Figure 9A), all or nearly all larval mortality was due to intrinsic constraints placed upon survival by food quality. However, much of the reduction in survivorship observed for some food quality/food quantity combinations (Figure 9) can be traced to increased larval life, which increases the fraction of total mortality contributed by extrinsic processes.

Genotype affected survival at the various food qualities. All combinations of egg sizes and respiration rates (growth

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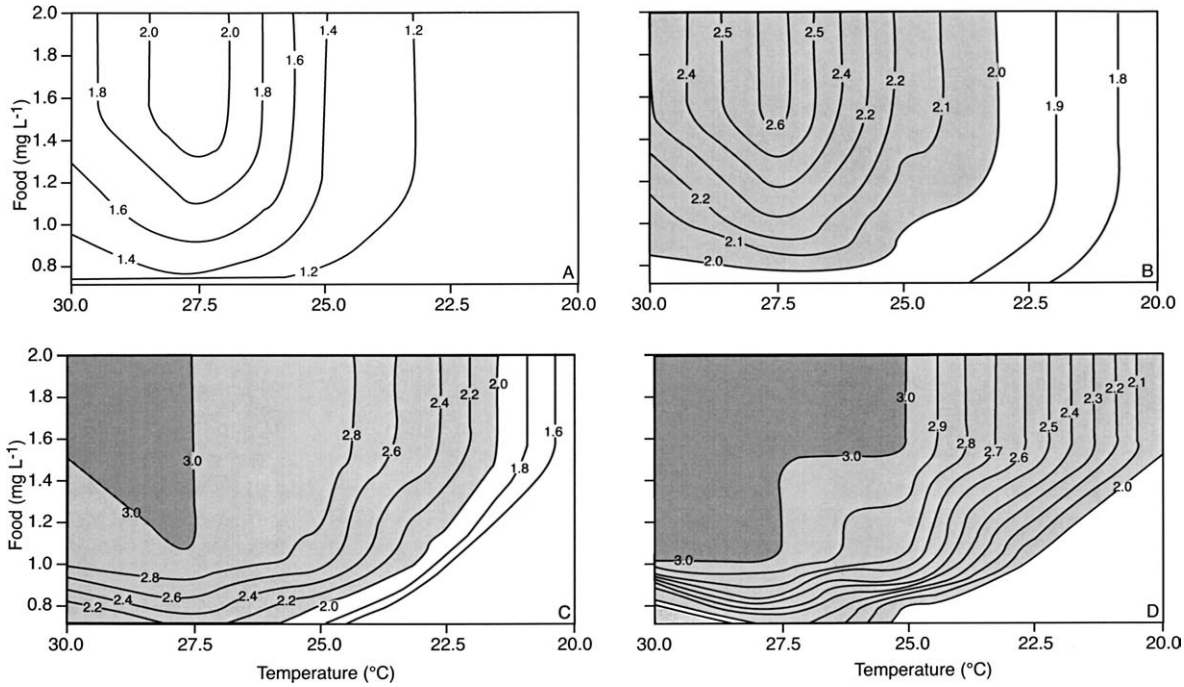

Figure 6. Average survival factor for (A) small eggs with high respiration rates, (B) large eggs with high respiration rates, (C) small eggs with low respiration rates, and (D) large eggs with low respiration rates, obtained from simulations that used a range of temperature and standard food ration combinations and a salinity of 25 psu. Contour values and shading are the same as in Figure 3. Note that the x-axis goes from high to low temperature.

efficiencies) did best at low protein-to-lipid ratios (Figure 10). The survival for small and large eggs with high respiration rates is not markedly different (Figure 10A, B). Successful metamorphosis is confined to low protein-to-lipid ratios and food concentrations above 0.5 mg L^{-1}. However, survivorship declined at the highest food concentrations and at the lowest protein-to-lipid ratios for larvae characterized by low respiration rates (Figure 10C, D).

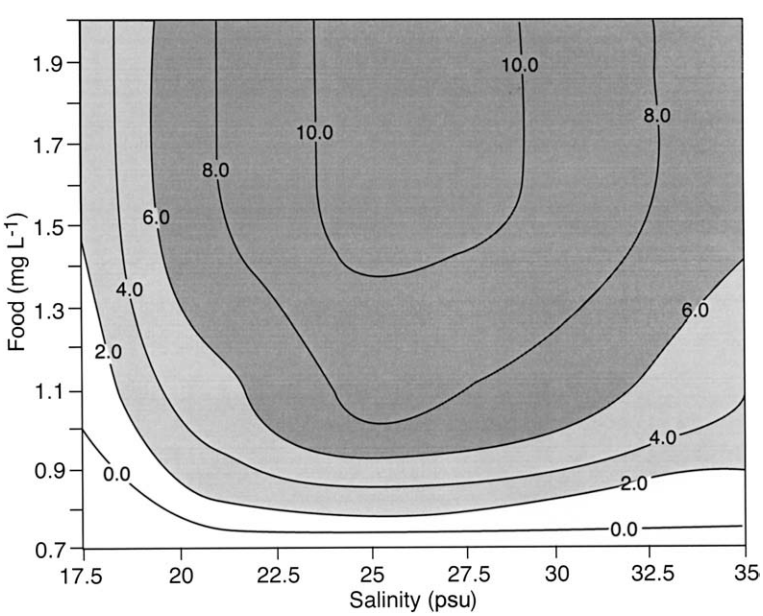

Figure 7. Percent of total cohort survival obtained from simulations that used a range of salinity and standard food ration combinations and a temperature of 27.5°C. Shading is the same as in Figure 2.

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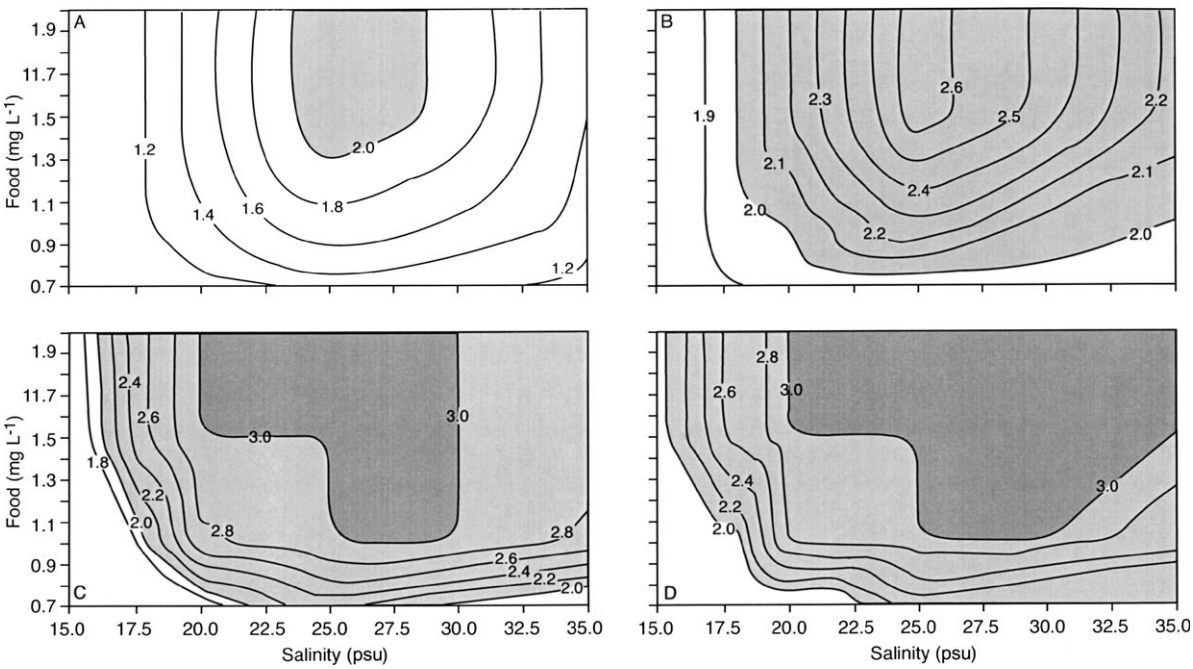

Figure 8. Average survival factor for (A) small eggs with high respiration rates, (B) large eggs with high respiration rates, (C) small eggs with low respiration rates, and (D) large eggs with low respiration rates obtained from simulations that used a range of salinity and standard food ration combinations and a temperature of 27.5 °C. Contour values and shading are the same as in Figure 3.

The same basic pattern was observed with the carbohydrate-to-lipid ratio; however, in this case the penalty on survivorship at high food rations and low carbohydrate-to-lipid ratios was increased over that observed at low protein-to-lipid ratios and the range of food qualities producing survival factors of 3 (no intrinsic mortality) was larger for larvae with low growth efficiencies than for larvae with high growth efficiencies (Figure 11). This represents the single most significant case where larvae from small eggs with low growth efficiencies were competitive with or had a competitive advantage over other larvae. The pattern of survival was more complex for variations in the carbohydrate-to-protein ratio (Figure 12). All larval genetic types were similar in having a survival factor near 3 at high food rations and high carbohydrate-to-protein ratios. However, at low carbohydrate-to-protein ratios, larvae with low growth efficiency had higher survival factors at high food ration.

Discussion

General characteristics of simulations

Year-to-year variability in recruitment is notable among finfish and shellfish, and the broodstock—recruitment relationships developed for these species reflect this variability (e.g. Hilborn and Walters, 1992). Variation in survivorship of larval cohorts is assumed to be a significant determinant of this year-to-year variation in recruitment. Larval survivorship is likely constrained by certain environmental conditions that directly limit survivorship. Those factors directly affecting the intrinsic ability to survive include the extremes of temperature, salinity, and food content. Less extreme environmental conditions may slow growth and, as the larval lifespan necessarily increases, impose increased cumulative predation losses on the larval cohort.

The limited ability to track larval cohorts in the field limits evaluation of the effect of environmental factors on larval survivorship. Nevertheless, studies, particularly of fish larvae, have implicated food availability as a critical factor affecting larval survival (Lasker, 1975, 1978; Lasker and Zweifel, 1978). Invertebrate larvae are likely to be similarly affected (Olson and Olson, 1989; Dekshenieks *et al.*, 1993, 2000; Fach *et al.*, 2002). The difficulties posed by direct observation of cohort survival in the field therefore require consideration of alternative approaches for investigating the factors affecting larval cohort survival. A larval growth model is one such option.

The model implemented here evaluates larval growth and survival at the level of basic biochemical constituents, and, as a result, allows investigation into the realm of variations in food quality. Changes in food quality, brought about by variations in the composition of phytoplankton, are likely to be common (Tester *et al.*, 1995; Philips and Badylak, 1996; Canuel and Zimmerman, 1999) and potentially as significant for larval survival as changes in food concentration. The reliability of the model predictions rests upon the

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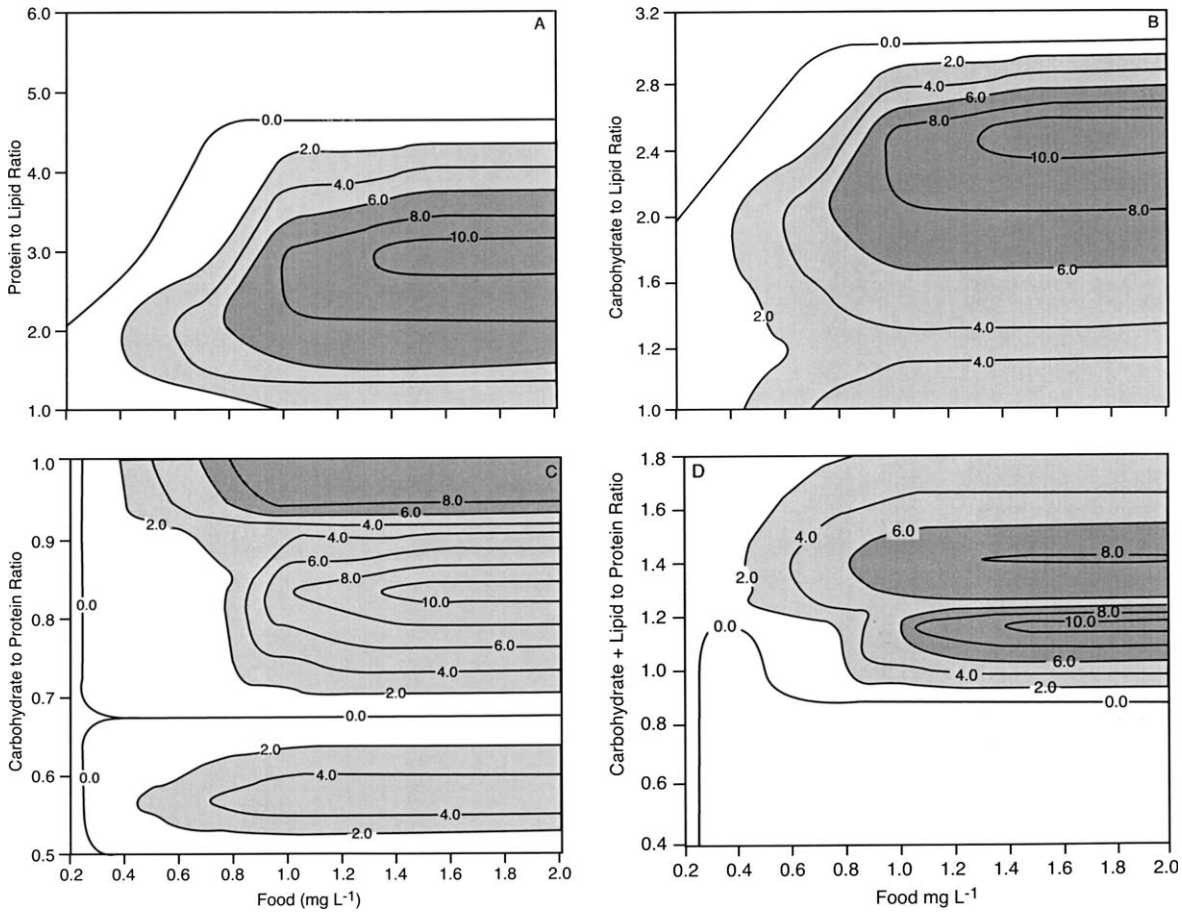

Figure 9. Percent of total cohort survival obtained from simulations that used a range of food qualities and food rations, a constant temperature of 27.5 °C, and a salinity of 25 psu. Food quality is given in terms of the (A) protein-to-lipid ratio, (B) carbohydrate-to-lipid ratio, (C) carbohydrate-to-protein ratio, and (D) carbohydrate + lipid-to-protein ratio. Shading is the same as in Figure 2.

parameterizations used to describe basic larval biochemistry and the sensitivity of the simulated larval growth to them (Bochenek *et al.*, 2001; Powell *et al.*, 2002). The greatest uncertainty in the model structure resides in the fate of protein when the food ingested has a high protein-to-lipid ratio. In the model, protein is only used for somatic growth, so that larvae fed a food composition rich in protein grow rapidly, but store insufficient lipid to sustain successful metamorphosis. The assumption that excess protein is not metabolized into other biochemical constituents or is dumped in some way leads directly to the low survivorships observed in these simulations. Such simulations underestimate survival resulting from food with lower protein ratios is observed in experimental studies (e.g. Helm *et al.*, 1973; Wikfors *et al.*, 1984; Whyte *et al.*, 1987; Thompson *et al.*, 1994, 1996; Garciá-Esquivel *et al.*, 2001) and similar results are reported for adult bivalves such as mytilid mussels (Hawkins and Bayne, 1991).

In addition, two other caveats must be considered. First, the outcome of many simulations hinges on the role lipid content plays in metamorphosis. The model structure is based on the assumption that variations in neutral and polar lipid dictate competency and metamorphosis success. Support for this assumption is provided by simulation results that agree with literature observations on metamorphosis, as described in Bochenek *et al.* (2001). However, little is actually known about the biochemical mechanism controlling metamorphosis success. Recent data (Garciá-Esquivel *et al.*, 2001) indicate that low larval survival was related to high protein use during metamorphosis, in agreement with the model assumption that relates higher lipid stores to increased metamorphosis success. Second, the simulations include a range of genotypes that are weighted in their importance based on a Gaussian distribution described by Equation (5). Little is actually known about genetic variability within cohorts. The range of genetic compositions implemented in the model is based upon experimental observation (Bochenek *et al.*, 2001), but the extent to which larval cohorts vary in the frequency distribution of selected genetic types is poorly known.

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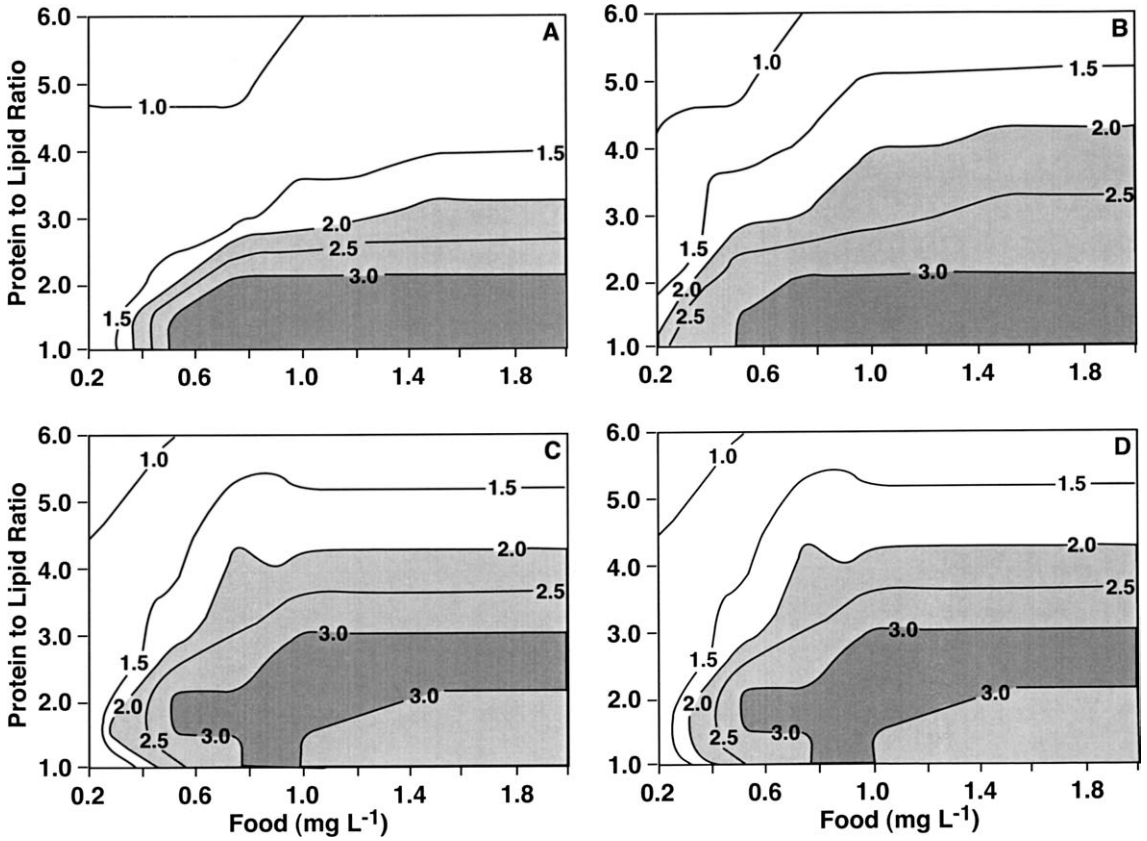

Figure 10. Average survival factor for (A) small eggs with high respiration rates, (B) large eggs with high respiration rates, (C) small eggs with low respiration rates, and (D) large eggs with low respiration rates obtained from simulations that used a range of food quality, defined by the protein-to-lipid ratio, and food ration combinations and a constant salinity of 25 psu and temperature of 27.5 °C, respectively. Contour values and shading are the same as in Figure 3.

Nevertheless, within the constraints imposed by the uncertainties described, the simulations provide a mechanism to assess the importance of environment, food quantity, and food quality on larval cohort survivorship and the likely importance of environmental change in determining the year-to-year variability in recruitment normally observed in bivalve populations.

Cohort survival

C. gigas larval survival is maximized, all else being equal, at temperatures and salinities of 25−30 °C and 25−30 psu (Figure 2), respectively, which are optimal values that are well documented in experimental studies (see Bochenek *et al.*, 2001). In comparison, the effect of food supply on simulated larval survival typically reaches saturating conditions at about 1.5 mg L^{-1} (cf. Figure 5) and survival declines at lower food concentrations. The food saturation level of 1.5 mg L^{-1} is much above the food content normally present as phytoplankton. However, Crisp *et al.* (1985) clearly documented this phenomenon and recent modelling studies have supported their conclusions (e.g. Dekshenieks *et al.*, 1993, 2000; Soniat *et al.*, 1998; Powell *et al.*, 2003). However, the food supply used in the simulations is well within the range of available food when food content is assessed using measurements of labile carbohydrate, lipid, and protein (Soniat *et al.*, 1998; Canuel and Zimmerman, 1999; Hyun *et al.*, 2001). Also, Crisp *et al.*, (1985) foresaw the need for this non-chlorophyll-dependent food to support larval growth. Thus, evaluating cohort survival at saturating food concentrations may be as significant as evaluating cohort survival under food-limiting conditions, because food-limiting conditions may not be nearly as common as once supposed based on estimates from chlorophyll measurements (Hirst and Bunker, 2003).

Variation in temperature and salinity away from optimum levels reduces larval survival, generally either because some larvae fail to metamorphose successfully or because increased larval lifespan increases predation loss. The intrinsic effects occur at the extremes of the environmental range (Figure 2). Over much of the range of environmental variation, the importance of predatory losses

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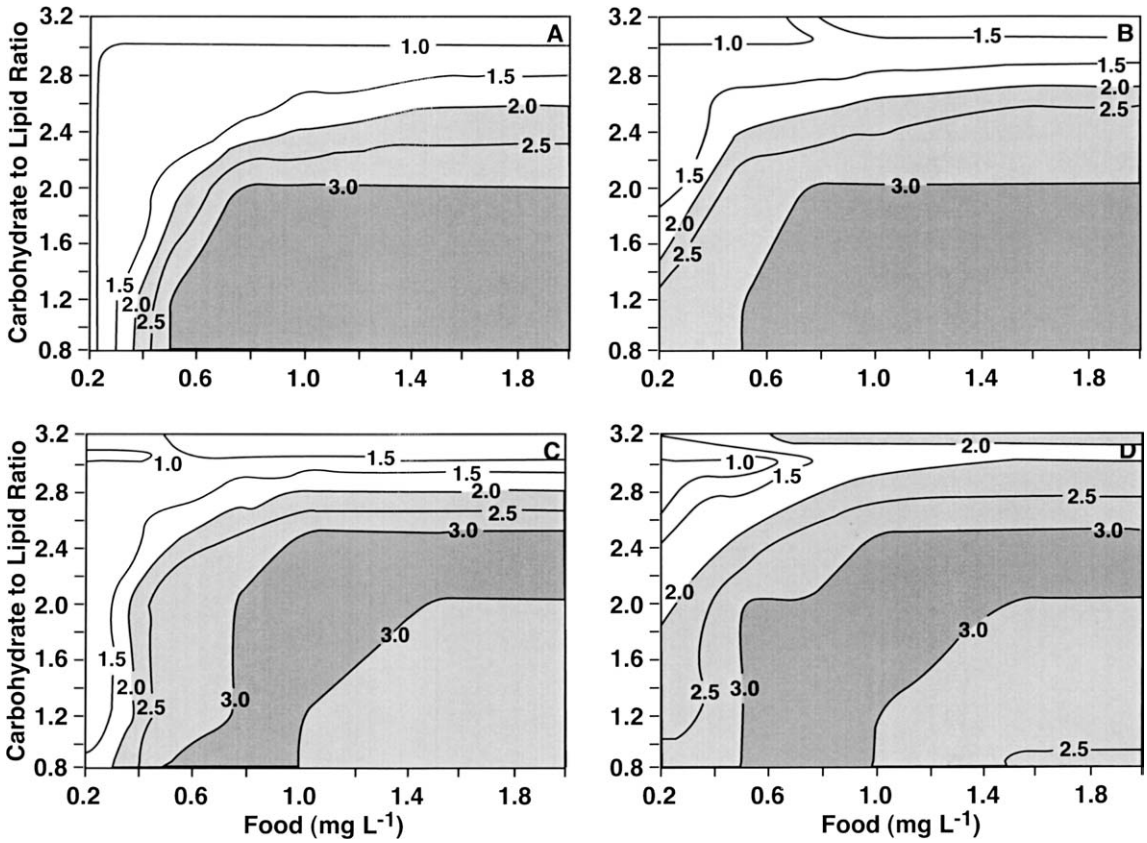

Figure 11. Average survival factor for (A) small eggs with high respiration rates, (B) large eggs with high respiration rates, (C) small eggs with low respiration rates, and (D) large eggs with low respiration rates obtained from simulations that used a range of food quality, defined by the carbohydrate-to-lipid ratio, and food ration combinations and a constant salinity of 25 psu and temperature of 27.5 °C, respectively. Contour values and shading are the same as in Figure 3.

in total mortality rises rapidly, so that determinants of the time required to become competent for metamorphosis determine the final degree of cohort survival (Figure 3). Larval lifespan is principally controlled by growth rate and the ability to store sufficient resources to sustain metamorphosis. The simple relationship between food supply and survival observed in the simulations is therefore expected. Variation in temperature and salinity from optimal conditions either increases respiratory losses or reduces ingestion, and so constrains the rate of growth and energy storage.

Certain temperatures and salinities improve survival at low food concentration. Higher temperatures increase ingestion or energy gain more than respiration or energy loss over a considerable temperature range (Figure 5). Variation in salinity tends to be relatively inconsequential at near-optimal temperatures (Figure 7). However, as temperature departs from the optimum, changes in salinity exert a constraint at all food concentrations, because growth rate decreases. Temperature negates the influence of salinity to a much greater degree than the reverse. Thus, a small increase in temperature, which improves larval survival, readily compensates for a larger change in salinity that would otherwise reduce survival. In contrast, a large change in salinity is required to compensate for the reduction in survival brought about by a small change in temperature, if compensation is possible at all.

Food quality

Food quality introduces another degree of complexity in that different food compositions produce widely varying survivals at the same food concentration. Certain food qualities limit survival because metabolic constraints result in death, such as a high protein-to-lipid ratio (Figure 9). Thus, intrinsic mortality can occur, and in fact be overwhelmingly important, under conditions of apparent environmental optimality based on salinity, temperature, and food concentration. Although some food combinations result in larval death during larval life, particularly early in life when feeding efficiency is low, in many cases intrinsic mortality occurs at metamorphosis when insufficient lipid is

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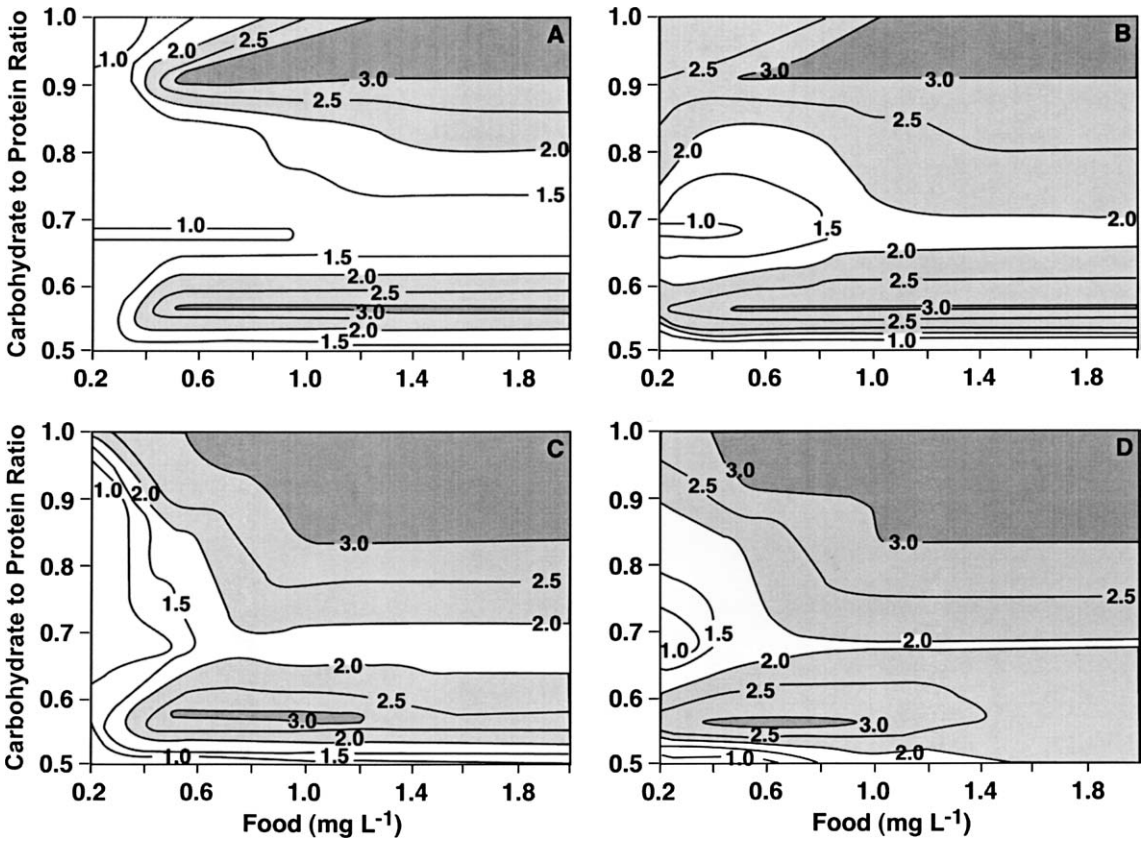

Figure 12. Average survival factor for (A) small eggs with high respiration rates, (B) large eggs with high respiration rates, (C) small eggs with low respiration rates, and (D) large eggs with low respiration rates obtained from simulations that used a range of food quality, defined by the carbohydrate-to-protein ratio, and food ration combinations and a constant salinity of 25 psu and temperature of 27.5 °C, respectively. Contour values and shading are the same as in Figure 3.

stored to successfully support metamorphosis. This occurs principally in diets that mismatch protein and lipid content. An implication of this result is that it is essential for somatic growth and lipid storage to occur in a uniform way. Otherwise, either growth rate will be too slow and the cohort will be lost to predation or too little lipid will be stored to support the reorganization of somatic tissue at the time of metamorphosis. As previously stated, the accuracy of this inference rests on the degree to which protein ingestion drives somatic tissue growth at all food compositions and on the lipid-dependent trigger used to initiate metamorphosis. Nevertheless, measured time-series of food quality which show temporal variations in constituent ratios at least as large as those used in the larval growth simulations (Soniat *et al.*, 1998; Canuel and Zimmerman, 1999; Hyun *et al.*, 2001; Versar, 2002) gives credence to the inference that the proportion of lipid and protein present in the food supply during larval life may substantially influence larval cohort survival.

Simulations indicate that food quality also influences growth rate and, accordingly, survival. Foods with insufficient protein directly reduce growth rate by limiting the rate at which somatic tissue can be produced. Foods with insufficient carbohydrate have the same effect on growth, but the resulting reduction in growth is due to the necessary use of other tissue constituents to support respiration. Increased lipid spares the carbohydrate need, but not the need for protein. As a result, the ratio of carbohydrate + lipid-to-protein may best describe the overall quality of the food (Figure 9). Simulations indicate that optimal food compositions are characterized by carbohydrate + lipid-to-protein ratios in the range of 1.1 to 1.4 over a range of food concentrations.

Low food supply restricts growth, but certain food qualities mitigate the expected decrease in survival brought about by a reduction in food concentration. Also, certain temperature ranges that permit an increased ingestion rate can compensate for low food concentrations. In the case of food quality, carbohydrate + lipid-to-protein ratios above 1.2 result in substantially improved survival at food rations below 1 mg L^{-1} (Figure 9D). For the most part, this relationship is driven by changes in intrinsic mortality. At lower carbohydrate + lipid-to-protein ratios, insufficient energy reserves are present to sustain the larva during

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metamorphosis and early in life when feeding efficiency is low.

Food qualities with extreme compositions containing too much or too little protein (Figures 10–12) reduce larval survival at saturating food concentrations. Too little protein results in slow growth and a longer lifespan, whereas too much protein impairs success at metamorphosis. However, genetic composition can partly compensate for the imbalance between growth and energy storage brought about by certain food compositions in high concentration. Thus, survival of the entire cohort may not be as affected as the survival of selected genotypes (Figures 9, 10).

The simulations show clearly that food quality can be as limiting to larval survivorship as can be food quantity, temperature, and salinity. Thus, the concept of a critical period, describing the need for larvae to have adequate food resources at certain times in larval development, should be expanded to include the concept of food quality.

Influence of genetics

Little is known about genetic variation in bivalve larval cohorts, and in particular about the influence of genotype on larval survival in the field. Genetic variation was included in the simulations by allowing variability in egg size and respiration rate; the latter being a surrogate for growth efficiency. Genetic variation in growth efficiency is well described and it may accrue from any number of processes including variations in respiration rate, protein turnover, assimilation efficiency or feeding efficiency (e.g. Garton, 1984; Koehn and Hilbish, 1987; Garton and Berg, 1989; Koehn and Bayne, 1989; Garton and Haag, 1991; Hawkins and Bayne, 1991). In terms of model parameterizations, variation in any of these processes produces essentially the same effect, because salinity and temperature control on larval growth is not resolved at the level of ingestion and respiration (e.g. Bochenek *et al.*, 2001).

The influence of genetics was inferred by identifying, through a series of simulations, the combinations of egg size and growth efficiency that permitted larvae to metamorphose successfully. For most environmental variables, including temperature, salinity, and food supply, larvae with high growth efficiency that began development as large eggs out-performed other egg size–growth efficiency combinations (Figures 6, 8). The influence of these three environmental variables was relatively simple through much of their range. Divergence from the optimum condition reduced growth rate by limiting ingestion or increasing tissue maintenance, which in turn limits lipid storage for metamorphosis and impacts the sufficiency of initial egg energy stores to provide the resources needed early in larval life when feeding efficiency is low. Thus, larvae from small eggs often failed to survive the first few days of life. Small eggs also resulted in longer lifespans and, consequently, higher predatory losses, for larvae surviving the first few days. Large eggs were relatively more successful because their larger energy stores provided a jump-start into larval life. These findings concur with the experimental findings of Gallager *et al.* (1986) and Gallager and Mann (1986).

Not surprisingly, optimal conditions of temperature, salinity, and food content are optimal not only because larval lifespan is minimized, thus minimizing predatory losses, but also because a wide range of genetic variants produces larvae that successfully metamorphose (Figures 6, 8). As conditions diverge from the optimum, an increasing number of egg size–growth efficiency combinations fail to support complete survival. Salinity, temperature, and food concentration, for the most part, affected genetic combinations that were initially confined to the small egg-high growth efficiency quadrant (quadrant 1 in Figure 1). As conditions continued to diverge from optimal, more egg size–growth efficiency combinations in quadrants 2 (large eggs, low growth efficiency) and 3 (small eggs, high growth efficiency) became affected. Normally, at the most extreme conditions in which any survival occurred, only larvae from large eggs with low growth efficiencies survived.

The same simple progression in survival did not occur with changes in food quality. Certain food qualities were most successfully used by selected genetic combinations of egg size and growth efficiency. For example, larvae with small eggs and low growth efficiency successfully metamorphose only under a limited range of salinities, temperatures, and food contents. However, larvae with low growth efficiency out-performed larvae with high growth efficiency at low carbohydrate-to-lipid ratios if food supply was high (Figure 11). Little difference was observed among the genetic combinations at food supplies of about 0.6–1.0 mg L^{-1}. Above a food level of 1.0 mg L^{-1}, larvae with low growth efficiencies had a selective advantage at low carbohydrate-to-lipid ratios. In the model, a surplus of lipid produced by these ratios resulted in the bypassing of the triggering method for metamorphosis, a lipid drop of a designated amount, when food was plentiful. Of course, the adequacy of this conclusion requires verification by experiment of the trigger for metamorphosis. Bochenek *et al.* (2001) discuss the implementation of the metamorphosis parameterizations used in the model in more detail.

Simulations varying the protein-to-lipid ratio followed a relatively similar pattern (Figure 10). Even the more complex interaction imposed by the ratio of carbohydrate-to-protein resulted in certain food compositions that favoured larvae with low growth efficiency (Figure 12). Thus, certain food qualities favoured larvae with low growth efficiency. These food compositions generally were produced by high carbohydrate- or lipid-to-protein ratios. Survival required the burning of carbon at a higher rate relative to the growth of somatic tissue, and this favoured larvae with low growth efficiency.

The dichotomy between food quality and the other environmental variables may be substantive in determining the range of genetic variation in a cohort. The simulations seem to suggest that the net influence of variations in

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temperature, salinity, and food content will be to compress genetic variation by uniformly favouring high growth efficiency and large eggs. The simulations with food quality provide evidence of a mechanism that would expand genetic variation, because variations in food quality favour a much broader range of genotypes. So, under certain conditions, low or high growth efficiency is a selective advantage, as are large eggs. A similarly unique selective advantage for small eggs was not evident in the scenarios tested with the larval growth model. This may be explained by the limited genetic range implemented in the model. Alternatively, the advantage of small eggs may be provided by the increased number of individuals spawned. The positive influence on survivorship that accompanies an increased number of eggs spawned is counterweighted by the extrinsic and intrinsic survival advantage of larger eggs.

Environmental instability

Simulations indicate that small changes in certain parts of the temperature/salinity/food range or small changes in food quality can result in large changes in larval survival. As an example, a change in food content from 1 mg L^{-1} to 0.5 mg L^{-1} effectively terminates larval survival (Figure 5). Time-series of phytoplankton standing stock show that variations on this order are routinely observed (e.g. Pennock, 1985; Jordan *et al.*, 1991; Cranford *et al.*, 1998; Sin *et al.*, 1999). Increasing the protein-to-lipid ratio from 3 to 4 reduces survival by 50% (Figure 9A). A 2 °C change in temperature varies survival by 50% (Figure 2). The response pattern for each of these variables is significantly non-linear, however, so that other environmental changes of equivalent magnitude produce only limited changes in larval survivorship. In addition, certain environmental changes reduce the impact of other environmental changes on larval survival. For example, an increase in temperature can counter the influence of a decline in salinity by minimizing a change in growth rate. The effect of a decline in food concentration can be minimized by an appropriate change in food quality. Thus, small changes in the environment can, under select circumstances, effect large changes in larval survival, whereas other changes, apparently of equivalent severity, have only a limited effect. This fact, coupled with the highly variable environment described by salinity, food quantity, and food quality criteria, may explain the substantially year-to-year variation in recruitment pattern and the apparently random nature of it.

Conclusions

Over the past several decades, larval growth models have become increasingly complex. These models, however, have utilized the standard processes governing energetics, including ingestion, assimilation and respiration, to simulate growth and survival. Such models have proved to be effective in addressing a wide variety of issues. However, the issue of food quality is not directly amenable to models of this sort. The influence of food quality is manifested through the biochemistry as the animal processes ingested carbon, whereas factors such as food quantity simply influence ingestion rate. The results of simulations using a biochemically based model to evaluate issues of food quality provide insights into how food quality affects cohort survival. Of particular importance is the fact that some diets counterweigh the negative effects of restrictions in food supply, in that lower rates of ingestion do not always result in equivalent decreases in cohort success. Some diets perform better in maintaining relatively high growth rates when ingestion rate is limited. Thus, the simulation of animals, like molluscan larvae, that are sensitive to changes in food content is improved by inclusion in the model of an underlying biochemical structure supporting the basic processes of ingestion, assimilation, growth, and respiration. The suggestion from a number of sources that oyster larvae require a relatively high food concentration for survival suggests that those variations in food quality that might permit survival at lower food concentrations might be particularly significant in increasing oyster larval survival in the field.

The results of the simulations suggest that greater scrutiny should be given to the metabolic fate of food components after ingestion over a range of food resource compositions. Certain observations, such as the impact of high protein diets and the influence of high lipid diets when food is plentiful, are based on assumptions about the fate of food constituents that are, as yet, not well verified. In addition, Strathmann *et al.* (1993) suggested that larvae can adapt morphologically to changing food availability. Such adaptations remain to be included in larval growth models and, if commonplace, would be significant in ameliorating some deleterious environmental changes. Moreover, adaptations at the biochemical level would also be significant. Evidence for such adaptations is not yet available. Thus, the complex interplay of temperature, food quantity, and food quality uncovered by the present simulations may only touch the surface of what is undoubtedly a complex milieu of adaptive responses to changes in the environment.

Much consideration has been given to the likelihood that variations in survival during planktonic life are responsible for the large observed variability in benthic recruitment. The results of simulations using a model that resolves larval performance at the level of the biochemical composition of the larva and its food supports the supposition that large variations in cohort survival can be explained by conditions encountered by planktonic larvae. Order of magnitude changes in survival are effected by relatively average variations in environmental variables such as temperature, salinity, and food supply. In addition, food quality appears to be a significant variable determining cohort success. The simulations strongly support the possibility that food quality is an important variable controlling cohort success and that variations in food quality potentially may be a key mechanism maintaining genetic variability in the larval population.

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