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Combined Effects of Changes in Temperature and Salinity on Early Stages of *Rangia Cuneata*[1,2]

Abstract—Embryos and early straight-hinge larvae of *Rangia cuneata* were subjected to a temperature change of 8°C in 5 seconds, a salinity change of 4‰ (5 to 1‰), and to the combination of both stresses. Embryos were relatively insensitive to the thermal shock but were killed by the salinity shock and by the combined salinity—temperature shock. Survival and growth of larvae were reduced by the salinity shock and by the combination of temperature and salinity shocks. The combination of the thermal shock and salinity change produced a mortality higher than the sum of the mortalities for either of the two factors alone.

Introduction

The use of estuarine waters for once-through cooling of a steam-electric power plant may result in damage to non-motile organisms that are forced to go through the plant cooling system (1). It is not generally recognized that the "entrained" organisms may be subjected not only to thermal and pressure changes and mechanical stresses but may also be subjected to a salinity change. In a number of power plants located in estuarine waters, the salinity of the intake water differs by several parts per thousand from that of the receiving water. The salinity difference results from the requirement that the cooling water intake must be located at some distance (or at different depths) from the discharge in order to avoid recirculation. As a consequence, there is a need for studies of the response of potentially entrained organisms to the combined stress of a thermal shock and a salinity change.

The availability of fertilized eggs during a broader study of the early life stages of the clam *Rangia cuneata* (Gray) and access to an apparatus designed to subject organisms to thermal shock permitted me to conduct an experiment on the sensitivity of embryos and larvae to the stress consisting of a thermal shock followed by a rapid salinity change.

Materials and Methods

Test organism: Rangia cuneata is a bivalve common in upper estuaries and low salinity embayments along the southeast and gulf coasts of North America (2). Although the adults can inhabit waters ranging in salinity from fresh through 15‰, development of the embryos is restricted to more stenohaline conditions (3).

Physical apparatus: A heat exchange device designed by Dressel (4) was adjusted to produce a temperature increase (ΔT) of 8°C to water of 28°C during its 5-second transit through the device. The apparatus consists of a 600 mm Graham condenser with a restrictive nozzle to decrease the flow rate to the 5-second passage time. Heated water is circulated through the condenser to obtain the desired ΔT.

Methods: Four-hour-old embryos and 24-hour-old straight-hinge larvae were separately subjected to (1) thermal shock, (2) salinity shock, (3) combined thermal-salinity shock, and (4) controlled conditions. Spawning, culturing, and subsampling techniques, and the combined effects of temperature and salinity on survival and growth of *Rangia cuneata* embryos and larvae are described by Cain (3). In the current experiment, embryos were obtained by spawning *Rangia* in water at 5‰ salinity at 28°C. Approximately 16,500 recently-fertilized eggs were transferred to each of 12 one-liter breakers in new water of 5‰ salinity and held four hours (to the ciliated stage) in a constant-temperature (28°C) bath.

Experimental conditions were imposed as follows: three samples were passed separately through the heat-shock apparatus, heated from 28°C to 36°C in 5 seconds and cooled from 36°C to 28°C over a 15-minute period; three samples were passed separately through the apparatus without added heat, collected on a 44 μm screen, and placed in water of 1‰ salinity; three samples of embryos were passed through the apparatus, heated 8°C, collected on a 44 μm screen, placed in 36°C water of 1‰ salinity and cooled to 28°C over 15 minutes; and, three samples of embryos were passed separately through the heat shock apparatus without added heat.

All samples, regardless of treatment, were returned to the 28°C constant temperature bath, and incubated for 20 hours, until they developed to the straight-hinge stage. Quantitative subsamples were taken by concentrating the straight-hinge larvae in a 250 ml round bottom flask, shaking, and withdrawing two-2 ml samples with a pipette.

Twenty-four-hour old, straight-hinge larvae were reared and tested. Eggs from the previous spawning were held in a 30-liter container at 28°C and 5‰

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until they developed to the straight-hinge stage. Approximately 24,000 larvae were placed in each of 12 beakers and subjected to the same procedure as in the embryo experiment. After this treatment the larvae were reared to setting size, which normally takes 6 days at 28°C. Quantitative subsamples were collected as before and 50 larvae were measured to the nearest 5 μm with an ocular micrometer in a compound microscope.

Results

The combined effect of thermal and salinity shocks are presented in Table 1. The 8°C ΔT in 5 seconds produced only a 7% increase in mortality over the control. The effect of salinity shock alone and temperature-salinity killed all embryos (Table 1A).

The combined effect of changes in salinity and temperature on survival of straight-hinge larvae was not as pronounced as its effect on the embryos. The 8°C–5 second thermal shock had a slight effect on larvae while the salinity shock caused a mortality of 45% over the control group (Table 1B). The combined stress resulted in a 70% mortality of the larvae. The effect of the thermal shock on the growth of larvae produced only a 5% reduction in growth (Table 1C). A 23% reduction in growth resulted from the salinity change alone, while the combination of thermal shock and salinity change had an effect of 27%.

TABLE 1

Effects of Thermal and Salinity Shocks on the Survival of Embryos and on Survival and Growth of Straight-Hinge R. Cuneata Larvae (Thermal Shock = +8°C, Salinity Shock = −4‰)

A. Survival of 4 hour old embryos to straight-hinge larvae.

		Control	Salinity Shock (ΔS)	Thermal Shock (ΔT)	ΔT & ΔS
Replicates	1.	266	1	257	0
	2.	260	0	245	0
	3.	267	0	236	0
Mean		264.3	0	246	0
Mortality		0%	100%	7%	100%

B. Survival of straight-hinge larvae after 6 days.

		Control	Salinity Shock (ΔS)	Thermal Shock (ΔT)	ΔT & ΔS
Replicates	1.	406	209	346	51
	2.	377	238	377	159
	3.	384	198	378	145
Mean		389	215	367	118
Mortality		0%	45%	6%	70%

C. Length (microns) of larvae at end of experiment (6 days).

		Control	Salinity Shock (ΔS)	Thermal Shock (ΔT)	ΔT & ΔS
Replicates	1.	158	120	148	112
	2.	157	119	146	115
	3.	148	117	147	111
Mean		155	119	147	113
Reduction in Growth		0%	23%	5%	27%

Discussion

Rangia embryos and larvae were relatively insensitive to the thermal shock of 8°C for 5 seconds. However, the 5 second exposure followed by a 15 minute return to the ambient temperature is a less severe thermal stress than that which most organisms would experience at a power plant.

The detrimental effect of reducing the salinity from 5 to 1‰ on embryos reaffirms the previous work showing 1‰ to be outside their salinity tolerance region. Although *Rangia* straight-hinge larvae are more tolerant to lower salinity concentrations, they nevertheless suffered considerable mortality with the rapid decline in salinity. The combination of the thermal shock and salinity change produced a mortality higher than the sum of the mortalities for either of the two factors alone. The effect of a gradual change in salinity (10–30 minutes), as would actually happen during the operation of a power plant, was not studied.

Future work on *Rangia* and other species should be conducted to determine the effects of other ranges of temperature-salinity combinations during all of the organism's early stages.

It has been shown by Calabrese and Davis (5) and others, that the early stages of molluscs are more sensitive to environmental conditions than adults. Successful recruitment to adult bivalve populations is dependent upon the survival and settling of the planktonic stages.

If salinity changes, either singly or in combination with heat, could cause mortality to a significant portion of the larval populations, they could be an important factor in power plant siting or in the arrangement of the cooling water canals.

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