Comparison of eastern oyster (*Crassostrea virginica*) and blue mussel (*Mytilus edulis*) filtration rates at low temperatures

Luc A. Comeau, Fabrice Pernet, Réjean Tremblay, Stephen S. Bates
and Angeline LeBlanc

Fisheries and Oceans Canada
Gulf Fisheries Centre
P.O. Box 5030
Moncton, New Brunswick
Canada, E1C 9B6

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by

Luc A. Comeau[1], Fabrice Pernet[2], Réjean Tremblay[3], Stephen S. Bates[1]
and Angeline LeBlanc[1]

[1]Fisheries and Oceans Canada, Gulf Fisheries Centre, P.O. Box 5030, Moncton, New Brunswick, Canada, E1C 9B6

[2] IFREMER, Avenue Jean Monnet, 34200 Sète, France

[3] Institut des Sciences de la Mer, 310 allée des Ursulines, Rimouski, Québec, Canada, G5L 3A1

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TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

ABSTRACT

Comeau, L.A., Pernet, F., Tremblay, R., Bates, S.S., and LeBlanc, A. 2008. Comparison of eastern oyster (*Crassostrea virginica*) and blue mussel (*Mytilus edulis*) filtration rates at low temperatures. Can. Tech. Rep. Fish. Aquat. Sci. 2810: vii + 17 p.

Eastern oysters (*Crassostrea virginica*) and blue mussels (*Mytilus edulis*) were collected in the Gulf of St. Lawrence at the northernmost distribution area of *C. virginica* and maintained in cold water (0°C, 4°C or 9°C) over a 63-day period. Filtration rates were periodically determined in closed chambers initially inoculated with 10,000 phytoplankton cells mL^{-1}. For *C. virginica*, filtration at low temperatures was clearly an exception: the percentage of the experimental population clearing the phytoplankton cells from the closed chambers declined from peak values of 50% at 9°C to null values (no animals filtering) at 0°C. For *M. edulis*, the percentage ranged from 100% at 9°C to 17% at 0°C. With respect to absolute filtration rates, *C. virginica* cleared significantly fewer particles than did *M. edulis*. Moreover, unlike *M. edulis*, *C. virginica* showed no adaptation to cold during the 63-day experiment. Together these results suggest that *C. virginica* is disadvantaged in terms of grazing on seasonal phytoplankton blooms, including toxic (domoic acid) blooms, which have historically occurred at low temperatures (< 9°C) in eastern Canadian waters. We caution, however, that further experiments are required to ascertain how changes in food quality or availability may influence feeding behaviour of *C. virginica* at low temperatures.

RÉSUMÉ

Comeau, L.A., Pernet, F., Tremblay, R., Bates, S.S., and LeBlanc, A. 2008. Comparison of eastern oyster (*Crassostrea virginica*) and blue mussel (*Mytilus edulis*) filtration rates at low temperatures. Can. Tech. Rep. Fish. Aquat. Sci. 2810: vii + 17 p.

Des individus de l'huître américaine (*Crassostrea virginica*) et de la moule bleue (*Mytilus edulis*) ont été échantillonnés dans le Golfe du St-Laurent, qui correspond à limite nordique de *C. virginica*. Les animaux ont été maintenus dans de l'eau à basse température (0°C, 4°C or 9°C) pendant une période de 63 jours. Les taux de filtration à l'intérieur de chambres fermées ont été mesurés périodiquement. La concentration de nourriture initiale dans les chambres était de 10 000 cellules de phytoplancton mL^{-1}. Dans le cas de *C. virginica*, la filtration à basse température était plutôt exceptionnelle : le pourcentage de la population expérimentale qui filtrait activement des particules de phytoplancton dans les chambres fermées atteint un maximum de 50% à 9°C et diminue pour atteindre des valeurs nulles (aucun individu ne filtre) à 0°C. Pour *M. edulis*, ce pourcentage varia de 100% à 9°C à 17% à 0°C. Quant aux taux de filtration absolus, *C. virginica* filtra significativement moins de particules que *M. edulis*. En plus, *C. virginica* ne démontra aucune adaptation au froid pendant la durée de l'expérience (63 jours), contrairement à *M. edulis*. Tous ces résultats suggèrent que *C. virginica* ne bénéficie pas des floraisons de phytoplancton saisonniers, incluant les floraisons toxiques (acide domoïque), qui ont lieu historiquement à basses températures (<9°C) dans les eaux de l'est du Canada. Cependant, d'autres travaux sont nécessaires afin d'assurer que des changements dans la qualité et dans la disponibilité de la nourriture n'influencent pas le comportement alimentaire de *C. virginica* aux basses températures.

INTRODUCTION

The eastern oyster, *Crassostrea virginica* (Gmelin 1791), has a remarkable distribution range of approximately 8,000 km in the Western Atlantic (Carriker and Gaffney 1996). It can be found in estuaries along the Brazilian coastline and northward through the Gulf of Mexico to the Gulf of St. Lawrence (GSL), Canada. Accordingly, the species is highly tolerant of extremes in ambient temperatures. At low tide, *C. virginica* can survive temperatures up to 49°C (Galtsoff 1964), whereas at northern latitudes it can freeze solid and recover at high tide (Loosanoff 1965).

The ability to survive at such extreme temperatures implies physiological adaptation, including the capacity to feed. For suspension-feeding bivalves, low temperature decreases food intake by acting on the animals' physiological rates, and also by changing the water's physical properties, particularly viscosity (Podolsky 1994). Few studies have examined the effect of cold on the feeding behaviour of *C. virginica* (reviewed by Shumway 1996), and only two have dealt specifically with very low temperatures. Both papers are rather dated and do not provide filtration rate data; rather, they present a detailed account of the flow rate of water through the mantle cavity, commonly referred to as the pumping or ventilation rate. Galtsoff (1926) concluded that no pumping activity occurred in *C. virginica* exposed to temperatures below 5°C. Loosanoff (1958) reportedly improved the laboratory apparatus for measuring pumping rates and found that some individuals—albeit a minority—ventilated at temperatures between 0°C and 5°C. Moreover, Loosanoff noted that a small number of individuals produced feces at these low temperatures, which led him to conclude that feeding is possible, although uncommon, in cold water. Above 5°C, the fraction of the experimental population ventilating (and presumably feeding) increased up to approximately 17°C, at which point all individuals became active. Therefore, based upon scarce information available in the literature, it appears that feeding activity is the exception below 5°C but becomes progressively more common as the temperature rises above 5°C.

Oysters in the Loosanoff investigation were from Long Island Sound (LIS), USA (Fig. 1). The 5°C thermal threshold for feeding reported by Loosanoff coincides with minimum water temperatures at that location. Figure 1 (inset) shows the ambient water temperature falling to approximately 5°C over roughly a two-month period during winter. However, *C. virginica* is widely distributed in the Western Atlantic, raising an intriguing question: does its lower temperature threshold for feeding change with latitude? In winter, water temperature falls to about 24°C in the Gulf of Mexico and to sub-zero values in the GSL, the northernmost distribution area of *C. virginica* (Carriker and Gaffney 1996). In the GSL, estuarine temperatures are stable over a narrow range between -1°C and 0°C for five consecutive months in winter; water temperature is also low (mean < 9°C) during autumn and spring when major phytoplankton blooms occur. Acute cold conditions may have led to the development of a northern strain of *C. virginica* characterized by a very low temperature threshold for feeding. Such adaptability would be consistent with the latitudinal-compensation theory, which predicts that individuals at higher latitudes compensate for temperature-associated slowing of metabolic processes by increasing physiological activity (Levinton and Monahan 1983; Lonsdale and Levinton 1985). Latitudinal compensation is often attributed to microevolutionary adaptation to local climates and has been firmly demonstrated for various species and physiological functions (Conover and Schultz 1995; Dittman 1997; Pörtner 2002; Kokita 2004; Sukhotin et al. 2006).

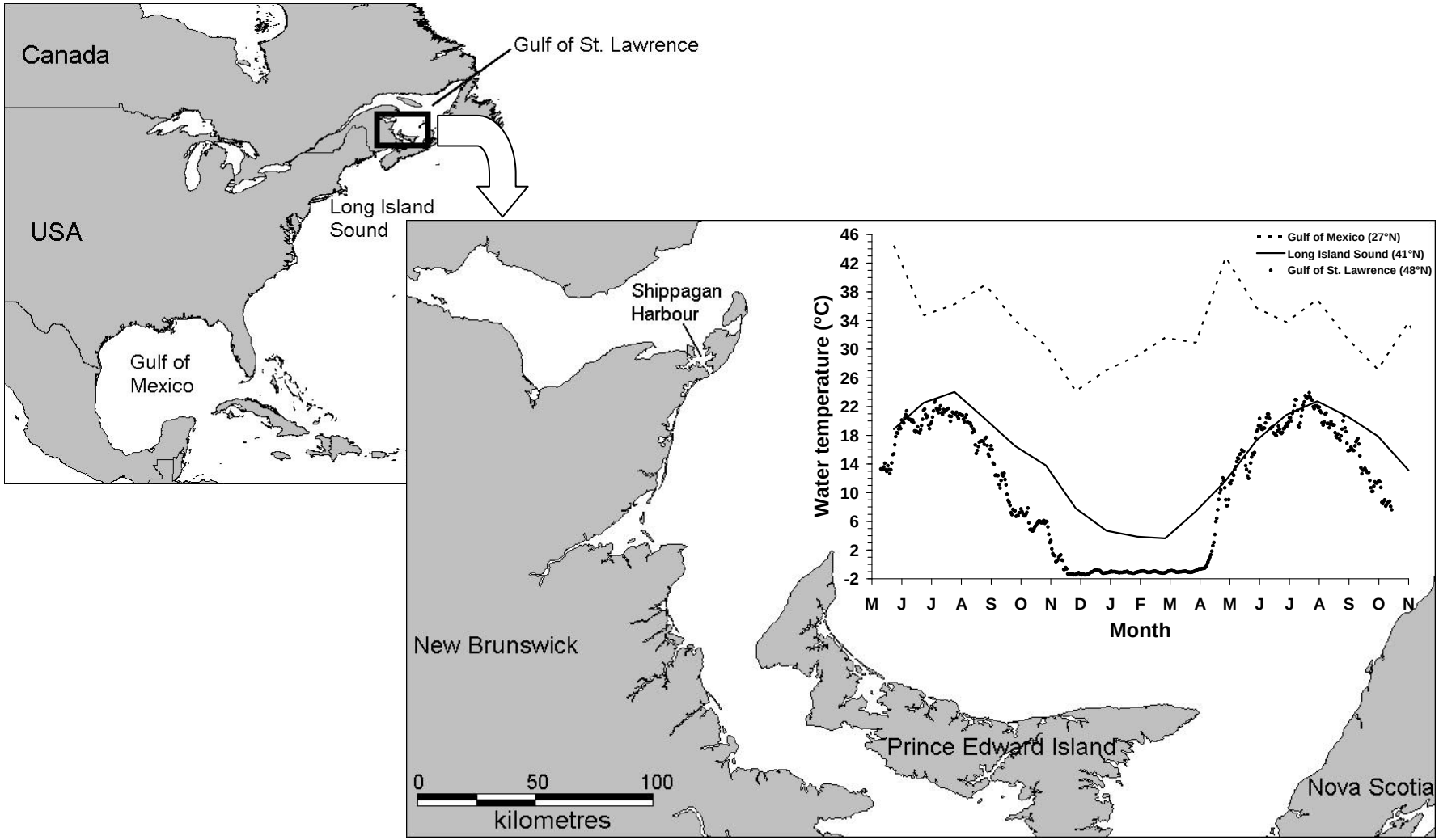

Figure 1. Map showing the three locations of *Crassostrea virginica* referred to in the text, and Shippagan Harbour, where the animals used in this study were collected. Water temperature time series are provided for the Gulf of St. Lawrence (DFO unpublished data, 2000-2001 daily means), Long Island Sound (NOAA National Data Buoy 44039, 2005-2006 monthly means) and the Gulf of Mexico (NOAA National Data Buoy 42020, 2005-2006 monthly means).

In this study, we collected *C. virginica* in the GSL and estimated its filtration rate in cold waters. Given that the animals were extracted from the northernmost (48°N) population, we postulated that they would be active below the 5ºC threshold previously reported for southern populations. The blue mussel *Mytilus edulis* (L.) was used during the present study as a benchmark for comparison. *Mytilus edulis* largely occupies the central part of North America, from North Carolina to Baffin Island (Gosling 1992). The GSL represents the northernmost area where the two species overlap in their distribution. *Mytilus edulis* was chosen as a benchmark for comparison because it is ubiquitous in the GSL, and there is convincing literature showing that *M. edulis* can consume food particles in near-freezing waters (Jørgensen et al. 1990; Loo 1992; Cusson et al. 2005; Kittner and Riisgård 2005). Finally, the comparison of *C. virginica* and *M. edulis* may provide practical information for the management of shellfish closures in the GSL. Harmful algal blooms (HABs), consisting of the domoic-acid-producing diatom *Pseudo-nitzschia multiseries*, have historically occurred in autumn (Bates et al. 1998; 2006; Trainer et al. 2008) or spring (Bates et al. 2002), when water temperature is low.

MATERIALS AND METHODS

Bivalves Collection and Maintenance

On November 15, 2004, approximately two months prior to the onset of ice formation, *M. edulis* and *C. virginica* were collected inside two neighbouring aquaculture leases in Shippagan Harbour, New Brunswick (Fig. 1). Water temperature at the two sites was 8°C and the salinity was 29 ppt. Animals were transported to the Coastal Zone Research Institute (CZRI), also located in Shippagan. Animals were tagged and carefully placed in one of two holding tanks (300 L) supplied with filtered (1 µm) and treated (UV and ozone) seawater. Water temperature was maintained at 9°C and salinity at 29 ppt. The natural photoperiod at 48°N was simulated using a light:dark (L:D) cycle starting with a 9:15 L:D period (November) and gradually changing to a 14:10 L:D period (April). Animals were fed a mixed suspension of live microalgae *Chaetoceros muelleri* (CHGRA) and *Isochrysis galbana* (TISO) every second day (2.0×10^4 cells mL^{-1}, 50:50 by cell number). Microalgal stocks were obtained from the Provasoli-Guillard National Center for Culture of Marine Phytoplankton (West Boothbay Harbor, Maine, USA).

Temperature Manipulations

On January 17, 2005, following a 63-day acclimation period at 9°C at the CZRI, the field-collected bivalves were randomly distributed amongst six experimental tanks. Each tank initially received six *M. edulis* and six *C. virginica* individuals. The experimental tanks were identical to the acclimation tanks and were supplied with the same source of filtered seawater (9°C, 29 ppt). However, each was equipped with its own chilling apparatus (1/2 or 1/5 HP chillers supplied by J&L Aquatics, British Columbia, Canada).

Following the transfer of animals, water temperature was lowered from 9°C to 0°C in tanks 1 and 2, and from 9°C to 4°C in tanks 3 and 4. This cooling was conducted at a rate of ~0.5°C d^{-1} over a 14-day period and was completed on January 31 (Day 0 of the experiment). Targeted temperatures of 0°C and 4°C were maintained until April 12, thereby simulating "cold" conditions over a 63-day period. In the two control tanks (numbers 5 and 6), water temperature was kept constant at 9°C. Temperature was closely monitored in all tanks using Minilog-TR data loggers (Vemco, Nova Scotia, Canada).

Standardized Filtration Rate

Filtration or clearance rate (*CR*) is defined as the volume of water cleared of suspended algal cells per unit time. Six animals of each species per tank were used for *CR* measurements (12 animals per temperature and species). Periodically (ca. biweekly), these animals were removed from their tank and placed individually in 600 mL acrylic chambers. These chambers were partially submerged in the animal's original tank to control temperature, and water from the tank was recirculated through the chamber. Following a 1-hour adaptation period in the experimental chamber, the water flow was halted and the chambers became static (Riisgård 2001). TISO was added at an initial chamber concentration of 10,000 cells mL^{-1}. TISO mixing was promoted by fine bubble aeration, introduced in a manner that minimized the resuspension of feces. Particle depletion within the chambers was monitored by counting suspended particles at the start of the incubation and subsequently at 10-minute intervals during a 1-hour period. Particles were counted by extracting water samples (10 mL) from the chambers and processing aliquots (100 µL) using a Beckman Coulter Counter Z1™ fitted with a 50-µm aperture tube. *CR* values reported here correspond to those obtained during the 10-minute interval that exhibited the greatest depletion of particles (Gilek et al. 1992; Cusson et al. 2005). This approach avoided the potential underestimation of *CR* due to particle levels falling below a critical threshold for valve opening (Newell et al. 2001; Riisgård et al. 2003) near the end of the incubation period. The 10-minute interval that showed the most depletion normally (80% of all cases) occurred during the first 30 minutes of the 1-hour incubation period. *CR* was calculated according to the formula

$$CR = \left[\left(\ln C_1 - \ln C_2\right) - S\right] \cdot V \cdot T^{-1},$$

where C_1 and C_2 are the particle concentrations showing the highest depletion rate during a 10-minute interval; S is an exponential sedimentation constant (0.0487 and 0.0591 for *M. edulis* and *C. virginica*, respectively) derived from reference chambers holding shells only (coefficient of variation for S was < 7% for shells of both species); V is the volume (600 mL) of water in chambers; and T is the time (10 minutes) elapsed between measurements. No mortality occurred during the course of the experiment. However, one mussel released gametes and was excluded from data analyses.

Statistical Analyses

The null hypothesis guiding statistical analyses was that feeding behaviour (*CR*) is unaffected by the factors *species* or *temperature*. Because *CR* measurements were repeated over five dates and measurements were consistently performed on the same individuals, the hypothesis was tested using repeated measures ANOVAs. For model building, time (*sampling day*) was identified as the within-subject factor, whereas *species* and *temperature* were labelled as between-subject factors. Individual tanks were considered as the unit of replication such that for each temperature treatment there were two replicate measurements taken at five different occasions. The tank mean *CR*, tank maximum *CR*, and the percentage of animals displaying positive *CR* values in each tank (i.e., percent feeding) were considered response (dependent) variables. Separate ANOVAs were performed for each of these response variables. Data were log+1 or arcsine/sqrt(x) transformed to achieve normality of residuals and homogeneity of variances. Eta-squared (η^2) values were calculated to determine the magnitude of the effects. Where significant effects were detected, *post hoc* multiple comparison tests were used to identify which means were different. Analyses were carried out using SPSS (SPSS Inc., Chicago, Illinois, USA).

RESULTS

Experimental Setting

Data on the shell length of animals are presented in Table 1. *C. virginica* and *M. edulis* measured 77–84 mm and 58–62 mm, respectively. The average shell length for each species was similar amongst the six experimental tanks ($P > 0.05$, Kruskal-Wallis). Table 1 also shows that the target temperatures were attained and were well replicated.

Table 1. Mean, standard deviation (SD) and sample size (*n*) for shell length and water temperature as a function of species and experimental tank.

	Shell length (mm)						**Temperature (°C)**	
	M. edulis			***C. virginica***				
Tank	**Mean**	**SD**	***n***	**Mean**	**SD**	***n***	**Mean**	**SD**
1	61	1	6	79	3	6	-0.3	0.3
2	58	2	6	82	4	6	-0.6	0.3
3	62	1	4	77	6	6	4.2	0.5
4	60	2	6	84	5	6	4.0	0.6
5	60	2	6	79	5	6	9.1	0.2
6	61	1	6	79	4	6	9.3	0.7

Crassostrea virginica *vs* Mytilus edulis

CR results are summarized in Table 2 for *M. edulis* and *C. virginica*. Means and associated error terms were derived from feeding individuals (non-feeding individuals were excluded). At 9°C, *M. edulis* cleared particles at an average rate of 1.82–2.90 L h^{-1} ind^{-1}. In contrast, the mean *CR* for *C. virginica* held at 9°C was comparatively low (0.05–1.21 L h^{-1} ind^{-1}). Similar differences between the two species were apparent at 4°C, but not at 0°C. Low *CR* sample sizes for *C. virginica* at 4°C and 0°C prevented a statistical comparison of means using a two-factor (*species* and *temperature*) ANOVA.

To increase sample sizes, we next considered the inclusion of non-feeding cases (*CR* = 0). In the bivalve literature, such cases are frequently attributed to stress associated with laboratory manipulations and therefore are often discarded from analyses (Cranford 2001;

Table 2. Mean clearance rate (*CR*) as a function of temperature and sampling day. "Sample size" indicates the total number of animals and the percentages that were clearing particles. Mean *CR* was derived from filtering animals only. Numbers in parentheses represent one standard error of the mean.

T	Day	*M. edulis*			*C. virginica*		
		Sample size		*CR*	Sample size		*CR*
(°C)		Total	% filtering	(L h^{-1} ind^{-1})	Total	% filtering	(L h^{-1} ind^{-1})
9	0	12	100	1.82 (0.88)	12	50	1.62 (0.74)
	14	11	73	1.93 (0.68)	12	17	0.87 (0.05)
	29	12	83	2.00 (1.01)	12	33	1.60 (1.21)
	43	12	100	2.49 (1.14)	12	17	0.69 (0.36)
	63	12	100	2.90 (1.30)	12	50	0.92 (0.36)
4	0	12	100	1.58 (0.59)	12	8	0.92 (n/a)
	14	12	92	2.40 (0.76)	12	8	2.68 (n/a)
	29	12	100	1.77 (0.67)	12	25	0.81 (0.37)
	43	12	92	2.09 (1.03)	12	8	0.14 (n/a)
	63	12	100	2.32 (1.46)	12	25	0.75 (0.87)
0	0	12	17	0.61 (0.13)	12	0	
	14	12	42	0.95 (0.38)	12	8	1.26 (n/a)
	29	12	50	1.46 (0.50)	12	17	1.27 (0.36)
	43	12	33	1.11 (1.09)	12	0	
	63	12	58	0.59 (0.26)	12	0	

Riisgård 2001). In the present study, however, it appeared that non-feeding animals were responding to the cold-water treatment. This assertion is supported by the significant reduction in the percentage of feeding *M. edulis* observed at 0°C compared with that at either 4°C or 9°C (Repeated Measures ANOVA, $P < 0.05$). For *C. virginica*, the proportion was significantly lower at both 0°C and 4°C compared to that at 9°C (Repeated Measures ANOVA, $P < 0.05$). These results linking non-feeding animals to controlled experimental conditions (temperature) provided a rationale for including non-feeding cases in our analyses.

We proceeded by evaluating the effects of species and temperature on tank-scale metrics, specifically, tank mean *CR* and tank max *CR*. The latter variable was assigned a null value when no animals where feeding. Results are summarized in Figure 2, and the corresponding ANOVAs are presented in Table 3. For *M. edulis*, the tank mean *CR* fell significantly at 0°C, whereas for *C. virginica*, the tank mean *CR* was similar at all investigated temperatures. A species effect was present—tank mean *CR* was significantly lower for *C. virginica* than for *M. edulis*, except at 0°C, where the two means were similar—but it was mediated by temperature (species × temperature, $P = 0.016$).

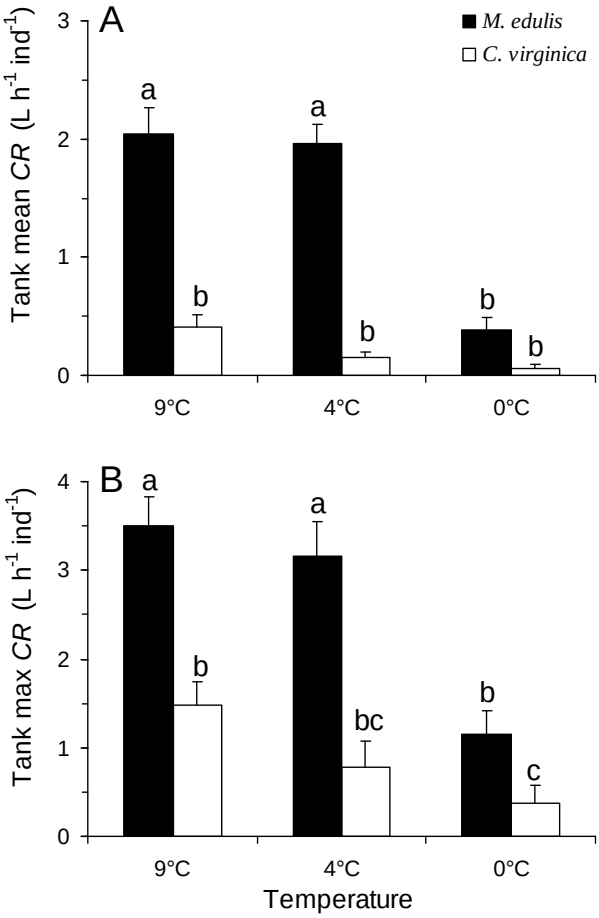

Figure 2. Mean (A) and max (B) clearance rate (*CR*) as a function of species and temperature. Error bars represent one standard error of the mean. Different letters indicate significant differences ($P < 0.05$).

Table 3. Mean clearance rate (*CR*) as a function of temperature and sampling day. "Sample size" indicates the total number of animals and the percentages that were clearing particles. Mean *CR* was derived from filtering animals only. Numbers in parentheses represent one standard error of the mean.

		M. edulis			***C. virginica***		
T	**Day**	**Sample size**		***CR***	**Sample size**		***CR***
(°C)		**Total**	**% filtering**	**(L h^{-1} ind^{-1})**	**Total**	**% filtering**	**(L h^{-1} ind^{-1})**
9	0	12	100	1.82 (0.88)	12	50	1.62 (0.74)
	14	11	73	1.93 (0.68)	12	17	0.87 (0.05)
	29	12	83	2.00 (1.01)	12	33	1.60 (1.21)
	43	12	100	2.49 (1.14)	12	17	0.69 (0.36)
	63	12	100	2.90 (1.30)	12	50	0.92 (0.36)
4	0	12	100	1.58 (0.59)	12	8	0.92 (n/a)
	14	12	92	2.40 (0.76)	12	8	2.68 (n/a)
	29	12	100	1.77 (0.67)	12	25	0.81 (0.37)
	43	12	92	2.09 (1.03)	12	8	0.14 (n/a)
	63	12	100	2.32 (1.46)	12	25	0.75 (0.87)
0	0	12	17	0.61 (0.13)	12	0	
	14	12	42	0.95 (0.38)	12	8	1.26 (n/a)
	29	12	50	1.46 (0.50)	12	17	1.27 (0.36)
	43	12	33	1.11 (1.09)	12	0	
	63	12	58	0.59 (0.26)	12	0	

A simpler statistical outcome was obtained for the second response variable, tank max *CR*. The species and temperature main effects were highly significant ($P < 0.01$) without interaction (species × temperature, $P = 0.146$). Tank max *CR* was substantially lower—by 66%, on average—for *C. virginica* than for *M. edulis*, and fell significantly at 0°C (compared to 9°C) for both species. Thus, *C. virginica* cleared fewer particles than did *M. edulis* within the range of investigated temperatures, and both *C. virginica* and *M. edulis* exhibited reduced feeding at 0°C.

A significant interaction between sampling day and species, which signals that the effect of time on *CR* was species-dependent, is another noteworthy result (Table 3). Figure 3 shows the estimated marginal means of *CR* plotted against sampling day for each of the two species. The predicted means of tank mean *CR* gradually increased with time for *M. edulis* but not for *C. virginica* (Fig. 3A). Similarly, the predicted means of tank max *CR* increased with time only for *M. edulis* (Fig. 3B); moreover, for tank max *CR*, the predicted means reached peak values before the experiment was completed.

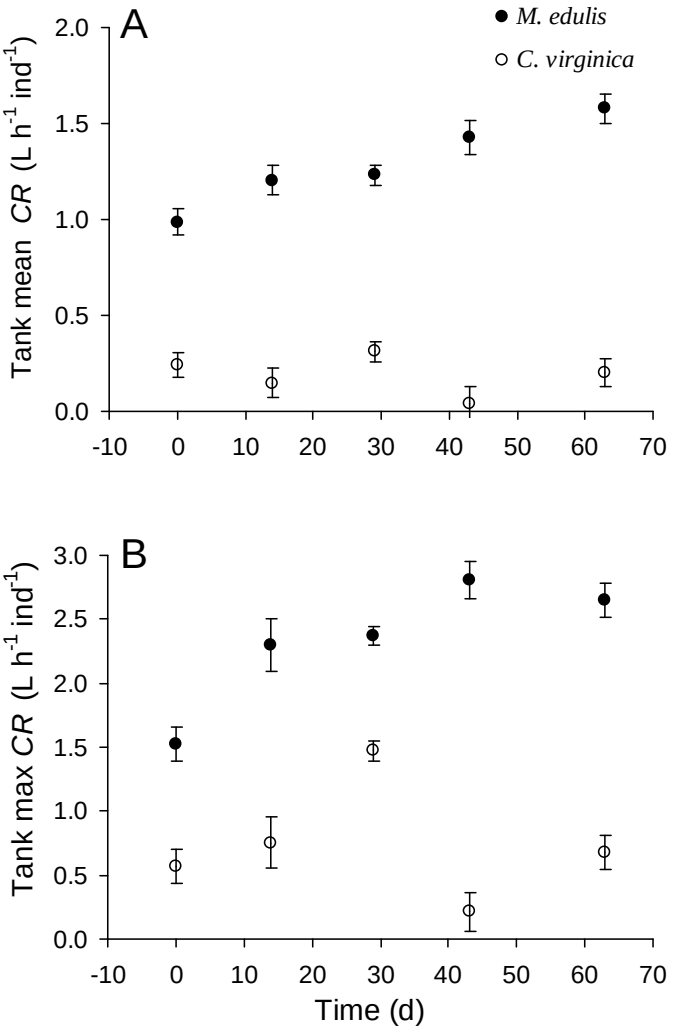

Figure 3. Mean (A) and max (B) clearance rate (*CR*) as a function of time. Values represent estimated marginal means. Error bars represent one standard error of the mean.

DISCUSSION

Feeding Behaviour of Crassostrea virginica

The feeding behaviour of *C. virginica* at low temperatures had previously been documented in only two older papers (Galtsoff 1926; Loosanoff 1958), and exclusively for individuals collected from a Long Island Sound (LIS) population. Here, we report that northerly Gulf of St. Lawrence (GSL) oysters behaved in much the same way as the LIS oysters in these earlier studies. Our data show a general trend, with the number of active animals decreasing with falling temperatures. The percentage of active *C. virginica* declined from peak values of 50% at 9°C to minimum values of 0% (no animals feeding) at 0°C (Table 2). Loosanoff (1958) found a similar reduction in activity, with percentage values falling from 44% to 2% within the same temperature range. Therefore, both studies support the assertion that *C. virginica* feeding is an exception in cold water.

It has long been established that a single *C. virginica* animal can filter up to 26 L h^{-1} under ideal conditions (Nelson 1938). In the present study, the maximum filtration rate for *C. virginica* was 2.7 L h^{-1} ind^{-1} at 9°C and 1.5 L h^{-1} ind^{-1} at 0°C. At the same temperatures in Loosanoff (1958), maximum pumping rates were 1.6 and 0.1 L h^{-1} ind^{-1}, respectively. The reason(s) for the lower rates in the Loosanoff study cannot be determined with certainty, although methodological differences between the two studies are potential explanatory factors. In the Loosanoff experiment, a rubber apron attached to the exhalent siphon was used to separate the exhaled water from the surrounding water, so that the former could be collected and measured. It has since been demonstrated that the rubber-apron technique creates mechanical disturbances and pressure gradients that inhibit pumping rates (Hildreth 1976; Famme et al. 1986). Thus, the laboratory apparatus used in Loosanoff (1958) may have resulted in an underestimate of pumping rates. In our study, we used an analytical approach to particle depletion that inherently selected the highest filtration rates.

Experimental oysters in the Loosanoff investigation were from LIS (41°N); those used in our investigation were collected in the GSL—the northernmost (48°N) distribution area of the species. Molecular genetics has thus far provided conflicting answers to the question of whether eastern Canadian oysters are genetically distinct from other populations found along the eastern coast of North America (Reeb and Avise 1990; Gaffney 1996; Hirschfeld et al. 1999; Hoover and Gaffney 2005). Here we found no supporting evidence for a latitudinal gradient in physiological function that related to feeding behaviour. Our data indicate that few northerly *C. virginica* individuals fed at low temperatures. For those few cases that displayed activity below 5°C, absolute feeding rates peaked at higher levels in the northerly GSL oysters than in the LIS oysters described in Loosanoff (1958). However, this observation may be linked to the methodological differences, as noted above, rather to evolved compensatory responses to cold conditions in the GSL.

Crassostrea virginica *vs* Mytilus edulis

Our null hypothesis—no difference in feeding activity between *C. virginica* and *M. edulis* exposed to cold waters—was tested using two clearance rate metrics: (1) tank mean *CR*, and (2) tank max *CR*. A "*species* effect" on tank mean *CR* was significant at 9°C and 4°C, but not at 0°C, indicating that our null hypothesis could not be rejected at the lowest experimental temperature. The tank mean *CR* can be viewed as an index of population-scale filtration rate, with the statistical outcome implying that, in extremely cold environments, population feeding rates are essentially the same for the two species. Although this conclusion seems valid for a two-month (63 day) exposure period (length of the experiment), it may not hold for longer exposures. As shown in Figure 3A, *M. edulis* (but not *C. virginica*) became progressively more tolerant to low temperatures during the course of the experiment. Given the linear nature of this relationship, it is reasonable to suppose that the tank mean *CR* for *M. edulis* at 0°C could have eventually become significantly higher than that of *C. virginica*. Note that estuarine temperatures in the GSL hover around 0°C over five consecutive months in winter (Fig. 1). There are conflicting reports in the literature on the ability of bivalves to acclimate to low temperatures (e.g., Widdows and Bayne 1971; Widdows 1976; Widdows 1978; Jørgensen et al. 1990; Riisgård and Seerup 2003; Petersen et al. 2003; Kittner and Riisgård 2005). In the present investigation, a controlled comparison of *M. edulis* and *C. virginica* suggests that *M. edulis* might be capable of cold adaptation over a more prolonged period (i.e., > 63 days).

Our second *CR* metric, tank max *CR*, may be regarded as the upper feeding rate in the experimental population. This metric was particularly sensitive to experimental conditions, given that any individual in the tank population could alter it. Moreover, since some individuals probably adapted faster than others, the metric likely captured the end physiological response earlier than did the tank mean metric. This interpretation is supported by the predicted tank max *CR*, which peaked after 43 days of cold treatment (Fig. 3B; *M. edulis*), whereas the predicted tank mean *CR* was still increasing after 63 days (Fig. 3A; *M. edulis*). A rapid response of any index tends to enhance the likelihood that significant differences between experimental groups will be found. In keeping with this logic, the max *CR* metric produced a consistent statistical outcome at each of the investigated temperatures: max *CR* was significantly lower—by 66% on average—in *C. virginica* than in *M. edulis*. Consequently, our null hypothesis was rejected for temperatures ranging from 0°C to 9°C. *Crassostrea virginica* at its northernmost latitude has a significantly lower filtration capacity than *M. edulis* during seasons when temperatures fall below 9°C.

Ecological Implications

In the present study, investigations were carried out at a relatively low food concentration (10,000 cells mL^{-1}). Since food availability is one of the main factors affecting feeding activity and filtration rate in filter-feeding bivalves (e.g., Hawkins et al. 1998; Cranford et al. 2005), further experiments should be performed at low temperature with different food concentrations (particularly with concentrations close to those that are naturally reached in the GSL during autumn and spring bloom events) to ascertain that oysters are not liable to increase their filtration

rates when the food availability increases, even at very low temperatures. In the interim, our data imply a major limitation for *C. virginica*—specifically, its inability to take full advantage of seasonal phytoplankton blooms in cold waters. In the GSL, these blooms occur during autumn and spring when estuarine waters are typically in the range of 4°C to 9°C (Fig. 1). Limited grazing during bloom events would explain the slow growth of individuals. For example, it takes a GSL oyster four to seven years to reach the legal market length of 76 mm, whereas oysters grown in the warm waters of the Gulf of Mexico may reach the same size in two years (Medcof 1961; Galtsoff 1964).

Finally, limited grazing during seasonal blooms also has implications for the accumulation of biotoxins by *C. virginica*. Domoic acid (DA) is a neurotoxin produced by certain diatom species of the genus *Pseudo-nitzschia* that are a food source for filter-feeding molluscs. High levels of DA have been sporadically found in *M. edulis* (Wright et al. 1989; Bates et al. 1998; Trainer et al. 2008), the sentinel species usually relied upon to trigger shellfish closures. While recognizing the suitability of *M. edulis* as a sentinel, relying on this species alone may unnecessarily restrict the harvest of other bivalves, such as *C. virginica*. In the spring of 2002, for example, DA reached 200 µg DA g^{-1} in *M. edulis* but remained undetectable in *C. virginica* at the same location in the southern GSL, or at trace levels (0.6-0.9 µg DA g^{-1}) elsewhere later in the spring (Bates et al. 2002). One plausible explanation for this difference is that *C. virginica* was not feeding—or feeding at very low rates—during the spring due to cold water temperatures. Our demonstration of a robust species effect on *CR* metrics in cold waters, with oysters feeding considerably less than mussels, supports this interpretation. Clearly, additional information on other mechanical and physiological processes at work in these oysters, including selection for algal species, assimilation of DA and depuration of DA at low temperatures, is required.

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