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FIELD PERFORMANCE AND QPX DISEASE PROGRESS IN CULTURED AND WILD-TYPE STRAINS OF *MERCENARIA MERCENARIA* IN NEW YORK WATERS

SOREN F. DAHL,[1] JOSHUA THIEL[2] AND BASSEM ALLAM[1]*

[1]*School of Marine and Atmospheric Sciences, 155 Dana Hall, Stony Brook University, Stony Brook, NY 11794-5000;* [2]*New York State Department of Environmental Conservation, Natural Resource Damages, 625 Broadway, Albany, NY 12233-1500*

ABSTRACT A field experiment was conducted to compare the performance of different hard clam (*Mercenaria mercenaria*) strains in local clamming waters of New York state. Experimental clams included a *Mercenaria mercenaria notata* seed obtained from a Florida broodstock, and 2 New York seed strains obtained from local hatcheries, including a cultured *M. mercenaria notata* strain and a first-generation "wild-type" strain. Quahog parasite unknown (QPX) was acquired by the Florida clams in less than 2 mo of a July deployment of grow-out cages. Prior field studies comparing susceptibility of northern and southern hard clam strains observed QPX acquisition after clams had overwintered in the field, raising the question that higher susceptibility observed in southern seed clams could be a result of poor adaptation to winter water temperatures. Our results show that the southern strain acquired QPX after the clams had only been exposed to the warmest period of water temperatures for this field site (22.3°C on average), thus excluding poor acclimation to winter temperatures as the main aggravating factor. In contrast, QPX was not observed until the second summer in the cultured New York (*M. mercenaria notata*) strain in which clam survival was high and infection prevalence remained minimal. The New York "wild-type" clams displayed good growth and did not acquire QPX at all, providing evidence for the potential utilization of local wild broodstocks to enhance the resistance of cultured strains. Histopathology observations offered further insights to infection dynamics, with early, light infections almost exclusively localized in mantle and gill tissues, clearly supporting the theory that these organs (predominately the mantle) are sites of acquisition for QPX infections.

KEY WORDS: *Mercenaria mercenaria notata*, wild type, clam strain, field trial, growth, survival, QPX, disease, infection dynamics

INTRODUCTION

Wild northern quahogs (=hard clams), *Mercenaria mercenaria* (Linnaeus 1758), from Raritan Bay, New York, serve as the source population for a valuable transplant fishery for the state. Transplant operations have been significantly curtailed because of the appearance of quahog parasite unknown (QPX) disease in that population during summer 2002 (Dove et al. 2004). No prior field trials concerning QPX disease have been conducted in New York, and therefore infection pressure of this protistan parasite in New York waters outside of Raritan Bay is unclear. The Peconic estuary system has been of particular concern because of its historical reception of clams from the transplant program for bacterial depuration and because the estuary is currently the focus of an ambitious bivalve aquaculture lease program (SCALP 2009).

Field trials investigating QPX disease have been documented in other states (Ford et al. 2002, Ragone Calvo et al. 2007) but did not include any clam strains from New York. These prior studies demonstrated that clam stock (host genotype) is an important determinant in clam susceptibility toward QPX. Disease dynamics in the field remain, however, largely unclear. For instance, Ford et al. (2002) observed increased intensity in late summer through autumn, then a decrease the following spring. In the study by Ragone Calvo et al. (2007), prevalence and intensity generally increased over time but had no clear pattern. Similarly, previous studies reported QPX disease in aquacultured clams that have been in the field for several months (9 mo or more) (Ragone Calvo et al. 1998, Smolowitz et al. 1998, Ragone Calvo et al. 2007), but the minimal time needed for disease development *in situ* remains imprecise.

*Corresponding author: E-mail: Bassem.Allam@stonybrook.edu

This article summarizes results of a field experiment that was conducted to compare the performance of 2 local hard clam seed strains in QPX enzootic waters of New York state (Raritan Bay). An additional *M. mercenaria notata* (Say 1822) seed obtained from a Florida broodstock known to be highly susceptible to QPX (Dahl et al. 2008) was included in this study to aid assessment of QPX infection pressure in Raritan Bay and in other clamming areas in the Peconic estuary. Particular focus was made to understand how quickly QPX disease can be acquired in a locally important enzootic area within Raritan Bay.

MATERIALS AND METHODS

Experimental Clams

Two different *M. mercenaria* hard clam strains were obtained from municipal shellfish hatcheries located on Long Island, New York. These included a hatchery-raised *M. mercenaria notata* strain used for municipal clam enhancement activities in New York state (NY *notata*) for several generations, and a first generation strain from a wild New York broodstock (NY white). A third strain was a hatchery-raised *M. mercenaria notata* variety from Florida (FL *notata*). All seed clams were within 10–12 mo of age and within a size range of 12–17 mm. The absence of QPX infection in experimental clams prior to deployment was confirmed using standard histological techniques (described later) and a minimum of 100 clams per strain was tested.

Field Deployment

Clams were deployed in 4 different experimental sites (Fig. 1). The first site was located in Raritan Bay (depth, 7 m) within an

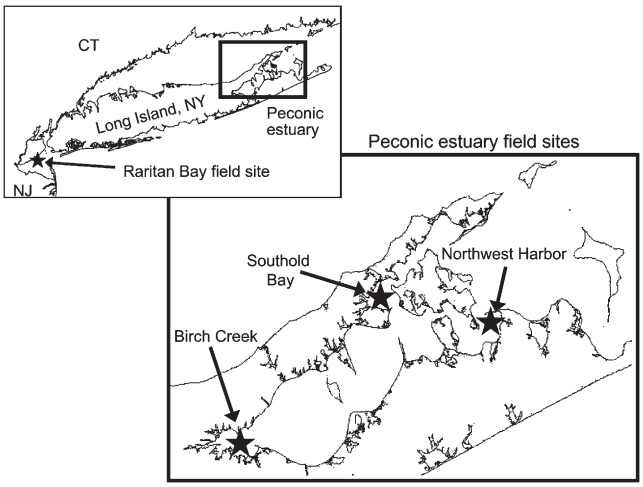

Figure 1. Map showing experimental field sites (stars) in Raritan Bay and the Peconic estuary (enlargement).

area that consistently tested positive for QPX since summer 2002. Birch Creek (depth, 1.5 m), Southold Bay (depth, 8 m), and Northwest Harbor (depth, 4 m) were located near clamming areas in the Peconic estuary that displayed occasional (Birch Creek and Northwest Harbor) or no presence (Southold Bay) of infected clams. In each site, 500 clams of a single strain were allotted to individual ADPI (ADPI Enterprises Inc. Philadelphia, PA) grow-out cages (OBC-1) 3/16-inch mesh (5 replicate cages for each strain per site). All 3 strains were deployed in Raritan Bay and Birch Creek. Because of a limited available quantity, only 3 replicates of the NY white seed were deployed in Birch Creek, and none were deployed in Southold Bay or Northwest Harbor. To keep the cages on the bottom, each side of the long axis was weighted with 1 segment of rebar. Individual cages were attached to a short line (leader) fastened to a longer line in a row (lines were cut from commercial lobster "pot warp"). The ends of each row were weighted down with cinder blocks or mushroom anchors to keep the deployment gear stationary. Global positioning satellite waypoints were recorded during deployment of each row of cages. Upon sampling, a grapple was dragged on the bottom to retrieve the lines. Original deployments occurred in the beginning of July 2004; final retrievals occurred in late October 2005.

Sampling

Sampling was conducted 3 times throughout the duration of the deployment and for a fourth time with the final retrieval (October 2005). The first 2 samples were planned for 2-mo intervals from deployment, starting in late August and again in October 2004; the third sample occurred the next summer after overwintering (June 2005). Unfortunately, the cages deployed in Birch Creek and Southold Bay were lost during the winter and thus were not sampled in 2005. During the first 3 sampling trips, each individual cage retrieved was subsampled for mortality counts, and 12 clams were taken for standard histological processing. From the first sample and again the following summer, 10 clams were taken from each cage for condition indexing (described later). Clam shell lengths were measured from condition indexing or from samples taken for histology. Daily growth rates were calculated as millimeters of shell growth per day for intervals between samplings. Underwater temperature data loggers (Onset Stowaway Tidbit, Onset Computer Corporation. Bourne, MA) were attached to the cages at original deployment and were either exchanged for another logger at the time of sampling and downloaded later, or downloaded in the field and redeployed immediately. During the final retrieval of clam cages, all remaining clams were counted to evaluate overall mortality, all remaining live clams were measured for length, and 30 clams from each cage were selected for histological analyses.

Condition Index

The condition index (CI) was calculated according to the suggested standard method (Eq. 6) in Crosby and Gale (1990):

$$\mathrm{CI} = \text{dry soft tissue weight (in grams)} \times 1,000/\text{internal shell cavity capacity (in grams)}$$

The formula is a modification by Hawkins et al. (1987) of the gravimetric techniques of Lawrence and Scott (1982). Clams were placed in an oven (60°C) and weighed over time until the dry weight had stabilized. They were then placed for 5 h in a benchtop muffle furnace set for 450°C. This process allowed the determination of ash-free dry weight, which was substituted for dry weight in the CI formula.

Histopathology

Sampled clams were shucked and placed in formalin (10% buffered) for preservation until dissection. A transverse slice of tissue roughly between 3 mm and 5 mm in thickness through the central region of the meat was made in an attempt to include visceral organs, as well as gill and mantle. Effort was taken to include tissue from the base of the siphon, where infections are commonly reported to initiate (Smolowitz et al. 2001). Tissue sections were placed in histology cassettes, embedded in paraffin, sectioned (thickness, 5–6 μm), and mounted on histology slides. Stained (Harris' hematoxylin for 2 min and Eosin Y for 1 min) slides were examined by light microscopy for presence of QPX. When QPX cells were discovered, the tissues infected and the infection intensities were quantified based on the number of QPX cells present on the histological section, and were recorded as follows: light (<10 QPX cells on the section), moderate (11–100 QPX cells), or heavy (101–1,000 QPX cells). A qualitative label was also applied to the observed distribution of QPX lesions: focal (localized in 1 well-defined area per tissue), multifocal (a few well-defined areas per tissues), and diffuse (profuse throughout major portions per multiple tissues).

Statistical Analysis

All statistical testing procedures were performed following methods described by Sokal and Rohlf (1995). QPX prevalence data and total mortality counts were analyzed for significant differences according to clam strain. Counts of QPX-infected and uninfected individuals from each histological diagnosis sample or the final total counts of dead and live clams were arranged in a 2-way, row-by-column contingency table and tested for independence of variables by means of the G-test through BIOMstat (Statistical Analysis for Biologists, version 3.3; Applied Biostatistics, Inc., Port Jefferson, NY). The first variable was classes of clam strain. The second variable was

either infection (with one class for infected and one class for uninfected) or viability (with one class for live and one class for dead). Counts were pooled from replicate samples. The frequency analysis program additionally carried out unplanned tests of all subsets of rows and columns in the row-by-column contingency table by Gabriel's simultaneous test procedure, which finds all maximal nonsignificant sets of rows and columns (i.e., a set that becomes significantly heterogeneous if any other row or column is added). CI and growth rate data were analyzed using SigmaStat for Windows (version 3.10; Systat Software, Inc., Chicago, IL). CIs were tested for significant differences of mean values for a particular clam strain across different field sites. Some of the CI data sets failed normality assumptions required for parametric testing of differences, and therefore the Kruskal-Wallis analysis of variance (ANOVA) on ranks test (Mann-Whitney rank sum test if only 2 samples) was conducted on pooled CI data. Daily growth rates were tested for significant differences of mean values for a particular field site according to clam strain as well as for a clam strain across different field sites using 1-way ANOVAs. Significant ANOVAs were followed by multiple comparison procedures: Holm-Sidak for growth rates (parametric) and Dunn's method for CIs (nonparametric). All results were considered significant at values of $P < 0.05$.

RESULTS

Shell Length and Growth Rates

Raritan Bay Site

Average sizes of FL *notata* and NY *notata* clams were similar at the beginning of the experiment, and both strains maintained similar increases in shell length throughout most of the deployment period (Fig. 2A) until the last measured increments between June 2005 and October 2005 (7.7 mm for NY *notata* compared with a 5-mm increment for FL *notata*). NY white clams were smaller at the beginning of the experiment than the other 2 strains, but had the greatest growth rate for each interval (Fig. 3A) and nearly doubled in length by the final sampling (Fig. 2A). Not surprisingly, all 3 clam strains had relatively poor growth rates during the winter (Fig. 3A), and most of the growth occurred during the first and second summer seasons. During the first summer, the growth rate was maximal in NY white clams, followed by FL *notata* and finally the NY *notata*. During the second summer, the growth rate of NY white clams remained highest, followed by NY *notata* and finally FL *notata*. Total growth rates measured throughout the entire deployment period were significantly different among different clam strains (ANOVA, $P = 0.0011$). Post hoc pairwise comparisons showed that NY white clams grew significantly faster than FL *notata* clams, whereas growth rates for NY *notata* was intermediary and not significantly different from those obtained in the other 2 strains (Fig. 3A).

Peconic Estuary Sites

In Northwest Harbor, FL *notata* and NY *notata* clams displayed similar increases in shell length during the first summer (Fig. 2B). After the October 2004 sampling, FL *notata* began to increase more substantially (Fig. 3B), resulting in a final average length that is 28% greater than the final average length of NY *notata* clams. However, as a result of significant mortalities at that site, there was only 1 cage of FL *notata* and 2 cages of NY *notata* with live clams left at the final measure, and therefore statistical comparisons could not be performed for the final interval. Clams deployed in Birch Creek and Southold Bay were lost during winter 2004 to 2005, thus data presented here represent information collected during the first 2 samplings (August 2004 and October 2004). In Birch Creek, only a small growth increment was noted among the FL *notata* clams (Fig.

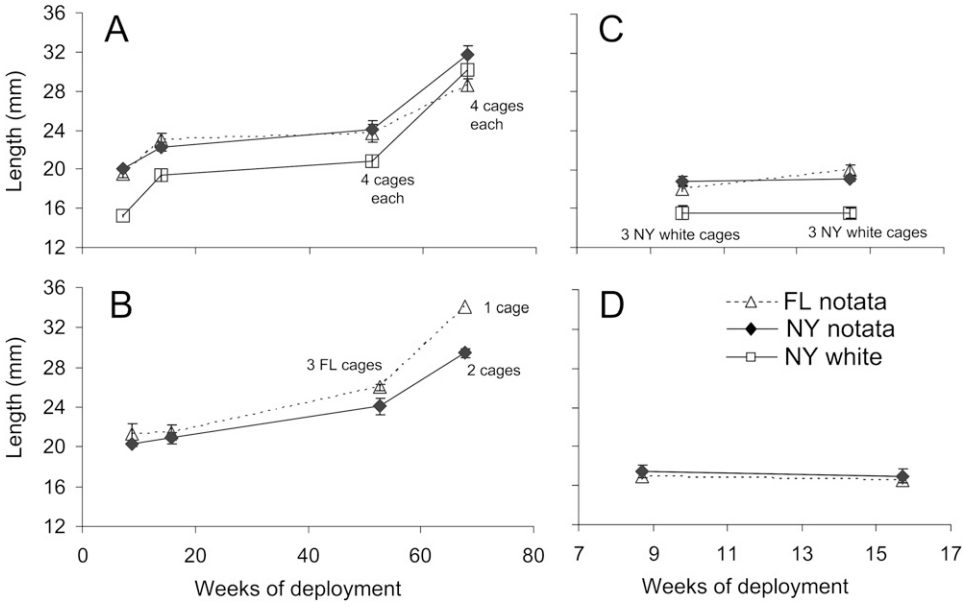

Figure 2. (A–D) Shell lengths (mean ± standard error) of clams deployed in Raritan Bay (A), Northwest Harbor (B), Birch Creek (C), and Southold Bay (D). Clams were sampled (10–12 clams per cage, 5 cages per strain unless noted otherwise) in August 2004 and October 2004 (all 4 sites), and June 2005 and October 2005 (A, B). Final samples in (A) and (B) represent a minimum of 85 clams per strain. NY *notata* and FL *notata: M. mercenaria notata* seed originating from New York and Florida, respectively. NY white: first-generation wild-type *M. mercenaria* seed from New York.

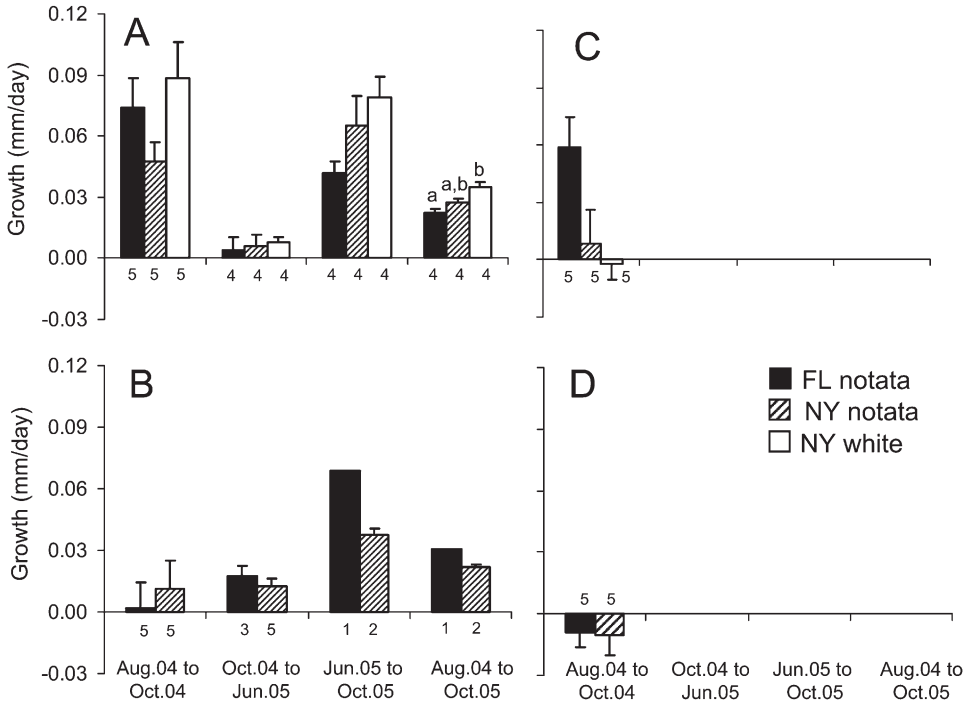

Figure 3. (A–D) Growth rates (mean ± standard error, 10–12 clams per cage, the number of replicate cages is given along the x-axis) of clams deployed in Raritan Bay (A), Northwest Harbor (B), Birch Creek (C), and Southold Bay (D). Different lowercase letters indicate significantly different growth rates among different strains (Holm-Sidak post hoc test, $P < 0.05$).

2C), and growth rates of the different clam strains were marginally significant (ANOVA, $P = 0.049$; Fig. 3C), but none of the pairwise comparisons were significant. In Southold Bay, neither the FL *notata* nor the NY *notata* clams showed any shell accretion (Fig. 2D). Calculated growth rates were slightly negative and were not significantly different between both strains (Fig. 3D).

Comparisons Among Different Sites

Growth rates measured during the first interval (August 2004 to October 2004) were compared for the same clam strain across different field sites. Growth rates for FL *notata* were significantly higher in Raritan Bay and Birch Creek compared with Southold Bay or Northwest Harbor (ANOVA, $P < 0.001$). NY *notata* growth rate means among different sites were marginally significant ($P = 0.047$), with only the values measured in Raritan Bay being significantly higher than those obtained in clams deployed in Southold Bay. Growth rates of NY white clams in Raritan Bay were significantly higher than those in Birch Creek ($P < 0.01$). Two-way ANOVAs (Sokal & Rohlf 1995) were also conducted to examine for significant interactions between sites and clam strains. Growth rates measured during the first interval were compared across all 4 sites for FL *notata* and NY *notata*, and all 3 clam strains for Raritan Bay and Birch Creek. No significant interactions resulted from either test.

Condition Index

CIs of different clam strains displayed a significant spatial pattern (Table 1). Seven weeks after deployment (August 2004), CIs obtained in Southold Bay were significantly lower than those obtained in Northwest Harbor (NY *notata*), and Raritan Bay and Birch Creek (FL *notata* and NY *notata*). In Raritan Bay, CIs displayed a significant increase ($P < 0.001$) over time for each clam strain (13% for NY white, 16% for FL *notata*, and 29% for NY *notata* clams). The opposite trend was observed in Northwest Harbor, with a significant decrease ($P < 0.001$) in CIs (38%) for both the FL *notata* and NY *notata* clams, which were significantly lower than those measured in Raritan Bay (Table 1). The condition of NY white clams was significantly lower in Birch Creek than in Raritan Bay.

Mortality

In Raritan Bay, cumulative mortality (Table 2) measured at the end of the experiment was higher for NY white and FL *notata* clams when compared with NY *notata* clams ($P < 0.001$). Similarly, mortality levels were higher in Northwest Harbor for FL *notata* clams when compared with NY *notata* ($P < 0.001$). FL *notata* endured significantly higher mortality in Northwest Harbor than in Raritan Bay ($P < 0.001$). The loss of deployed clams in Birch Creek and Southold Bay did not allow the presentation of total mortality in these sites, and data presented here represent information collected during the October 2004 sampling. In Birch Creek, mortality levels were highest in NY *notata*, followed by FL *notata* and finally NY white clams ($P < 0.01$). In Southold Bay, FL *notata* clams had significantly higher mortality than NY *notata* ($P < 0.001$).

QPX Prevalence, Intensity and Distribution in Clam Tissue

QPX was not detected in clams sampled from either of the 3 deployment sites in the Peconic estuary (Birch Creek, Southold Bay, and Northwest Harbor). Diagnosis of the histology

TABLE 1.

Condition index of clams (mean ± standard error; 10 clams per cage, the number of cages is as in Fig. 3) collected in August 2004 (all sites) and June 2005 (Raritan Bay and Northwest Harbor).

Field Site and Date	FL *notata*	NY *notata*	NY white
Raritan Bay, August 2004	120.61 ± 2.12[a]	124.49 ± 2.9[a]	130.24 ± 3.28
Birch Creek, August 2004	122.7 ± 2.73[a]	132.19 ± 0.48[a]	117.34 ± 0.95
Northwest Harbor, August 2004	130.36 ± 28.32[a,b]	135.32 ± 14.25[a]	Rank sum test, $P < 0.001$
Southold Bay, August 2004	109.59 ± 7.41[b]	110.19 ± 6.9[b]	
	ANOVA on ranks, $P < 0.001$	ANOVA on ranks, $P < 0.001$	
Raritan Bay, June 2005	137.63 ± 2.66	160.64 ± 13.74	147.57 ± 6.75
Northwest Harbor, June 2005	80.66 ± 0.48	89.93 ± 1.0	
	Rank sum test, $P < 0.001$	Rank sum test, $P < 0.001$	

Different lowercase letters designate a significant difference among sites for each strain (Dunn post hoc test on Ranks or Mann-Whitney Rank Sum test, $P < 0.05$).

samples taken at 7 wk (August 2004) and then 14 wk (October 2004) after deployment in Raritan Bay revealed QPX only in the FL *notata* clams (1.7% and 8.3%, respectively; Fig. 4A). QPX prevalence increased substantially in the FL *notata* clam samples taken after 51 wk (June 2005) to 47.9% and remained high (51.2%) in the final sample. Except for the first sample, all the FL *notata* samples had significantly higher QPX prevalence (G-test, $P < 0.01$) than both New York clam strains. The first infection was detected among the NY *notata* clams after 51 wk, resulting in a prevalence of 2.1%, which remained low in the final sample (2.5%). QPX infection was not observed in any of the NY white clams. Statistical analysis of the prevalence data for the final sample demonstrated significantly higher prevalence for NY *notata* clams when compared with NY white clams (G-test, $P = 0.04$).

The first QPX-positive FL *notata* clam (7 wk postdeployment) displayed a light focal infection in the mantle (Fig. 4B). The positive FL *notata* clams after 14 wk displayed mostly moderate infections, with focal infections restricted to the mantle, multifocal infections including siphon tissues, and the single heavy infection additionally displaying QPX in the visceral mass. The majority of QPX infections in the 51-wk FL *notata* sample were light (48%) or moderate (39%), and the remaining positive clams (13%) were heavy infections. In this sample, QPX was observed strictly in pallial tissues in all clams that displayed focal infections (48%)—often in the gills, but in the mantle and siphon tissues as well. QPX was also observed in the visceral mass in all but one of the multifocal infections (35%) and in all of the diffuse infections (17%). All the heavy infections displayed diffuse or multifocal lesions. A range of infection severity, distribution, and tissue combinations was represented in the final FL *notata* sample (67 wk, October 2005). Compared with the prior (51-wk) sampling, the proportion of light infections decreased as heavy infections increased, and reached nearly 25% each. Moderate infections represented the remaining half of the cases (51%). The proportion of focal infections decreased nearly in half in the final sample (from 48% at 51 wk to 25% at 67 wk) as the proportion of diffuse infections increased noticeably (17–39%). The first QPX-positive NY *notata* clam (51 wk) had a light focal infection in the siphon. In the final NY *notata* sample, 1 clam had a light focal QPX infection in the gills, 1 clam displayed a moderate focal infection at the junction between the mantle and the siphon, and another had a moderate focal infection in the foot.

Temperature Data

Temperature data were available for all sites between July 2004 and October 2004 (Fig. 5), and throughout the entire experiment only in Raritan Bay and Northwest Harbor. Temperature range was similar between Southold Bay (15.1–24.6°C) and Raritan Bay (15.7–24.8°C). Northwest Harbor had an approximately 2°C greater range (13.7–24.8°C), whereas Birch Creek displayed an approximately 5°C greater range (12.8–27.3°C). Compared with an average of the 3 Peconic temperatures, Raritan Bay remained 0.5°C cooler from mid July to mid September and 1°C warmer from mid September to mid October (Fig. 5). In general, the Peconic estuary sites appear to fluctuate greater and more frequently than Raritan Bay; this is especially noticeable in the Birch Creek plot.

TABLE 2.

Cumulative mortality (mean ± standard error) for each clam strain at each deployment site.

Clam Strain	Raritan Bay July 2004 to October 2005	Northwest Harbor July 2004 to October 2005	Birch Creek July 2004 to October 2004	Southold Bay July 2004 to October 2004
FL *notata*	68.0 ± 4.05[a]	78.2 ± 5.9	13.45 ± 2.83[a,b]	20.6 ± 2.31
NY *notata*	59.05 ± 3.3[b]	60.89 ± 10.54	18.38 ± 4.11[a]	10.61 ± 1.46
NY white	68.9 ± 3.02[a]	G-test, $P < 0.001$	7.57 ± 4.14[b]	G-test, $P < 0.001$
	G-test, $P < 0.001$		G-test, $P < 0.01$	

Raritan Bay and Northwest Harbor samplings cover up to October 2005; Birch Creek and Southold Bay cover up to October 2004. Different lowercase letters designate a significant difference among strains within a site (G-test, $P < 0.05$).

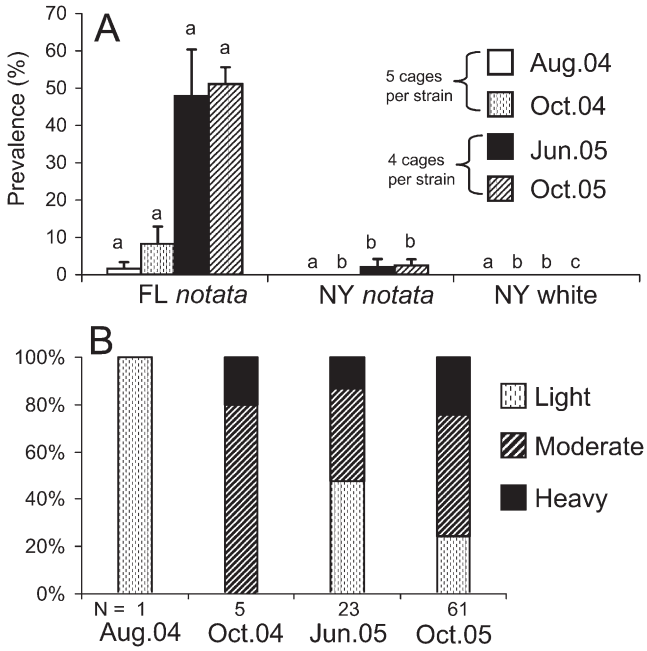

Figure 4. (A) QPX disease prevalence (mean ± standard error) in clams sampled from the Raritan Bay deployment (12 clams per cage except for October 2005 when 30 clams per cage were processed). Different lowercase letters designate significantly different prevalence among different strains sampled on the same date (G-test, $P < 0.05$). (B) Proportion of infected FL *notata* clams with different QPX disease intensities (the number of infected clams is given along the x-axis).

The temperature range through the entire deployment was almost 3°C greater in Northwest Harbor (range, –1.5–26.9°C) than in Raritan Bay (range, –0.2–25.5°C). A divergent colder trend is observed in Northwest Harbor when compared with Raritan Bay in the fall and winter (average difference of 1.4°C from mid September 2004 to mid March 2005). In spring and summer, there was only a slightly warmer trend in Northwest Harbor (average difference of 0.2°C from mid March 04 to mid September 2005).

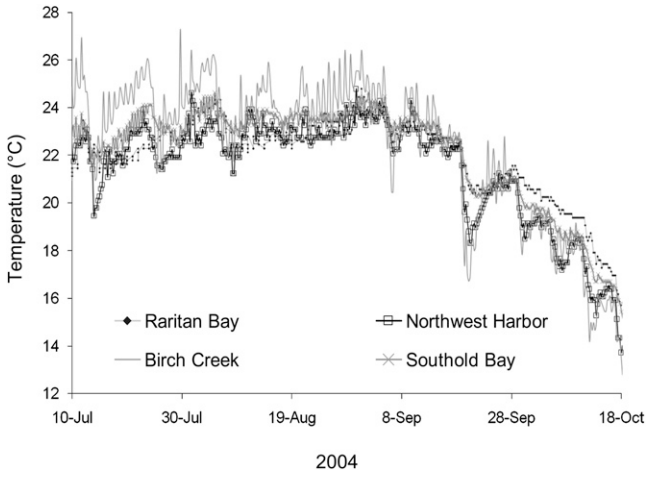

Figure 5. Water temperature recorded by data loggers deployed with clam cages. Plots are restricted to show only the period of time that data are available from all four field sites.

DISCUSSION

This study aimed at comparing the performance of locally cultured and wild-type hard clam strains (NY *notata* and NY white) in QPX enzootic Raritan Bay, New York. A second objective of the study was to evaluate QPX disease pressure in clamming areas in the Peconic estuary. The southern (Florida) hatchery-raised clam strain was deployed in addition to the New York strains to increase the chance of QPX detection based on the previously reported high susceptibility of southern clam stocks to QPX (Ford et al. 2002, Ragone Calvo & Burreson 2002, Ragone Calvo et al. 2007, Dahl et al. 2008), aiding potential sensitivity in discerning between clam resistance to infection as opposed to lack of QPX infection pressure. QPX disease is often reported in cultured clams, which has created concern that culture practices may increase the risk of infection. Clam stocking densities have been a suspected risk factor because they are often much higher than found in natural populations (Ford et al. 2002, Ragone Calvo & Burreson 2002). The stocking density of clam seed allotted to the grow-out cages in this study (1,000/m^2) was higher than previous QPX studies, but still well within the range of planting densities applied within aquaculture practices (550–1,650/m^2) for clams in that size range (12–17 mm) (Castagna 2001). This application was intended to monitor acquisition of infection and disease dynamics, high mortalities were to be expected, and ample individuals were used to ensure desired samples could be obtained. Use of juvenile seed to assess QPX infection pressure was validated in results from the Raritan Bay deployment. QPX was not histologically detected in samples from the Peconic estuary. This is a favorable outcome, providing consolation for concerns of a potential QPX epizootic in the Peconic estuary, especially when considering the historical role of receiving waters for the depuration of Raritan Bay hard clams.

Use of a highly susceptible hard clam seed strain allowed for rapid disease development, facilitating the study of *in situ* disease dynamics. The first histology sample of FL *notata* clams was taken from Raritan Bay after 7 wk of deployment, and a QPX-positive clam was discovered, representing the quickest acquisition of QPX reported *in situ*. Previous studies described QPX disease in hard clams that have been in the field for 9 mo or longer (Ford et al. 1997, Ragone Calvo et al. 1998, Smolowitz et al. 1998, Ragone Calvo et al. 2007). Earlier field studies have also reported clam strains from southern hatchery origins displaying greater susceptibility to QPX disease than northern clam strains after clams had overwintered in the field (Ford et al. 2002, Ragone Calvo & Burreson 2002, Ragone Calvo et al. 2007). Researchers suggested this disparity could be a consequence of the southern strains being poorly adapted to winter water temperatures in the northern field sites (Ragone Calvo & Burreson 2002, Ragone Calvo et al. 2007). In our study, the Raritan Bay deployment was initiated in July, the first sample was in August, the second was at the beginning of October, and both samples tested positive for QPX despite the fact that those periods cover the warmest water temperatures of the year in Raritan Bay (average, 22.3°C). This new information strongly supports an alternative hypothesis of genetically based susceptibility in southern (Florida) strains compared with northern stocks (New York, in our case) is in agreement with laboratory transmission experiments reported previously (Dahl et al. 2008).

High frequency of QPX infection prevalence in mantle and gill tissues in previous reports (Ragone Calvo et al. 1998, Smolowitz et al. 1998, MacCallum & McGladdery 2000), as well as in their own study, led Ford et al. (2002) to suggest that those tissues "are the portals of entry for QPX" (p. 34). Throughout our study, the early-stage infections were consistently found in pallial organs (mantle and gills), providing further evidence that these organs represent the site of infection initiation. In the field study of Ragone Calvo et al. (2007), mild infections were mostly localized in mantle tissue, and more severe infections tended to be multifocal. The results of our study are remarkably similar, because the light focal infections were consistently found in a pallial organ, whereas heavier and more diffuse infections observed QPX in the visceral mass as well.

Indication of infection seasonality has not been clear from prior field observations. QPX-related mortalities were highest in late summer in Massachusetts (Smolowitz et al. 1998). A seasonal survey conducted in Atlantic Canadian provinces found the highest prevalence in August samples (MacCallum & McGladdery 2000). In the current study, QPX acquisition and initiation of infection seems to occur during early summer and increases in disease severity up through autumn. For instance, the light infection observed in FL *notata* clams in August 2004 appears to have progressed into moderate and heavy infections that fall (October 2004). A similar scenario is observed for the NY *notata* clams starting during the second summer (June 2005) of deployment. Interestingly, light infections reappeared in the FL *notata* clams the following year (June 2005), likely representing another round of QPX acquisition. The final FL *notata* clam samples taken that fall (October 2005) also revealed an increased percentage of heavy infections. This rudimentary pattern of QPX infections is evident in several years of data from our monitoring of wild hard clams in Raritan Bay (Liu et al. 2008, Allam unpub.).

Results of growth rates and CIs demonstrated a clear influence of field site on the performance of each clam strain. A generalized influence seems unilateral across the clam strains, because good values for growth and condition where observed in Raritan Bay. In addition, there were no significant interaction effects as interpreted from the results of the 2-way ANOVAs. Good clam performance in Raritan Bay is not surprising given that this area maintains the most productive hard clam population in New York state. Alternatively, clam performance was very poor in Southold Bay, where "negative" growth rates were measured (Fig. 3D), probably as a result of sampling error or selective mortality of larger clams. In June 2005, both clam strains deployed in Northwest Harbor displayed a significant decrease in their CI compared with the prior year. The causes of this reduction are unclear, but it may be worth noting that heavy mortalities in both clam strains were seen during June 2005 and October 2005 samplings.

In general, the FL *notata* clams displayed some good instances of growth and relatively good condition, yet had high mortalities in most sites and an overwhelming difference concerning QPX disease prevalence. These findings are congruous with the study by Ragone Calvo et al. (2007), especially in terms of the significantly higher mortality and QPX prevalence for their southern clam strains (e.g., Florida and South Carolina) versus the northern strains tested. Survival of NY *notata* clams was high in most field sites with relatively good growth rates including the greatest CI increase in Raritan Bay. NY white clams had the lowest mortality in Birch Creek yet poor growth and condition there, whereas in Raritan Bay they had significant growth and good condition but higher mortality than the NY *notata*. It may be relevant to note that even though all efforts were made to obtain clams of each strain that were the same size, the NY white clams tended to be closer to the lower size selection limit of 12 mm.

Despite the marginal difference observed in terms of disease prevalence among both New York stocks, the fact is that the NY *notata* strain displayed less resistance to QPX than their "wild" counterpart in Raritan Bay. Differences in resistance to infection between the 2 stocks could be a collateral effect relating to differences among selection processes. The NY white clams are first-generation wild-type and not subject to the selection processes of the cultured *M. mercenaria notata*, and culturists may focus on favoring specific traits that subsequently results in an unintended physiological trade-off in other traits. For instance, our NY *notata* seed has been used for aquaculture for several generations and is generally assumed to be characterized by fast growth, although in Raritan Bay, the NY white clams actually had the best growth rates. Something in the Raritan Bay environment could have reduced the growth potential of the other strains but did not inhibit the NY white clams in the same regard. QPX disease likely decreased growth performance of more susceptible clams. The striking decrease in growth performance of FL *notata* measured during the second summer (Fig. 3A) compared with the first summer coincided with, and probably resulted from, an increased disease burden. Previous studies diagnosing hard clams from the field found heavily infected clams to be smaller and showed reduced growth when compared with clams with little to no infection (Smolowitz et al. 1998, Ford et al. 2002). Especially when considering the 2 New York strains tested, a better genotype environment match could also be the case even though no statistical "interaction" was found (2-way ANOVAs). Ragone Calvo et al. (2007) found that "particular stocks responded better to certain very local conditions" (p. 115). The fact is that the NY white clams showed great potential for field applications, particularly with regard to QPX disease resistance, but also in potential for growth proven in Raritan Bay.

Observations of *in situ* dynamics from this field trial reinforce the notion that infections are initiated in the pallial organs (Ford et al. 1997, Smolowitz et al. 1998, Smolowitz et al. 2001, Ford et al. 2002). A few weeks were sufficient for susceptible, naive seed clams to acquire infections, which became severe less than 4 mo after deployment. Findings also clearly demonstrate that differences in QPX susceptibility, observed in previous field trials (Ford et al. 2002, Ragone Calvo & Burreson 2002, Ragone Calvo et al. 2007), are based on hard clam genotype likely resulting from a lack of selection for resistance, and not primarily contingent on winter temperature stress. These findings corroborate results of QPX susceptibilities associated with clam genotype from our experimental transmission trials (Dahl et al. 2008) that were conducted under consistent warm water temperatures (20–21°C). Performance of the NY white clams illustrates the potential utilization of wild broodstocks in enhancing resistance of cultured strains. Future investigations should focus on the understanding of biological bases of disease resistance in northern stocks, and functional genomics tools are currently being developed to address these issues.

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